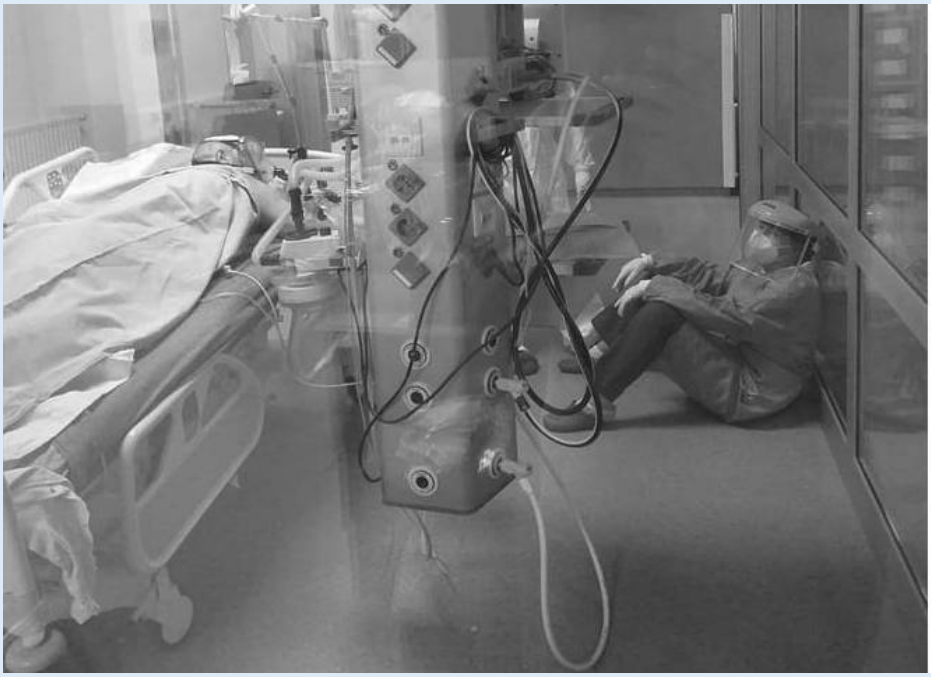
Glasnik hemičara i tehnologa Bosne i Hercegovine
Bulletin of the Chemists and Technologists of Bosnia and Herzegovina



57

December, 2021.

Univerzitet u Sarajevu - Prirodno-matematički fakultet, Sarajevo
University of Sarajevo - Faculty of Science, Sarajevo

Glasnik hemičara i tehnologa Bosne i Hercegovine
Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

57

December, 2021.

Univerzitet u Sarajevu - Prirodno-matematički fakultet, Sarajevo
University of Sarajevo - Faculty of Science, Sarajevo

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

Print ISSN: 0367-4444
Online ISSN: 2232-7266

Zmaja od Bosne 33-35, BA-Sarajevo
Bosnia and Herzegovina
Phone: +387-33-279-918
Fax: +387-33-649-359
E-mail: glasnik@pmf.unsa.ba
glasnikhtbh@gmail.com

EDITORIAL BOARD

Faculty of Science Sarajevo
Zmaja od Bosne 33-35, BA-Sarajevo
Bosnia and Herzegovina
Phone: +387-33-279-904 (Administration)
+387-33-279-911 (Executive Editors)
Fax: +387-33-649-359

E-mail: glasnik@pmf.unsa.ba
glasnikhtbh@gmail.com

Dejan Milošević
Department of Physics, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Željko Jaćimović
Department of Chemical Technology, Faculty of Meallurgy and Technology, University of Montenegro, Podgorica, Montenegro
Ljudmila Benedik
Department of Environmental Sciences, "Jožef Stefan" Institute, Ljubljana, Slovenia
Meliha Zejnilagić-Hajrić
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Tidža Muhić-Šarac
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Sabina Gojak-Salimović
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Ismet Tahirović
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Danijela Vidic
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Mustafa Memić
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Andrea Gambaro
Department of Environmental Sciences, Informatics and Statistics, University Ca' Foscari of Venice, Venice, Italy
Institute for the Dynamics of Environmental Processes - National Research Council (CNR-IDPA), Venice, Italy
Dragana Đorđević
Centre of Chemistry - IChTM, University of Belgrade, Belgrade, Serbia
Aida Šapčanin
Department of Natural Sciences in Pharmacy, Faculty of Pharmacy, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Jože Kotnik
Department of Environmental Sciences, "Jožef Stefan" Institute, Ljubljana, Slovenia
Lucyna Samek
AGH University of Science and Technology, Faculty of Physics and Applied Computer Science, Krakow, Poland
Angela Maria Stortini
Department of Molecular Sciences and Nanosystems, University Ca' Foscari of Venice, Venice, Italy
Ivan Spanik
Institute of Analytical Chemistry, Slovak University of Technology, Bratislava, Slovakia
Mirjana Vojinović Miloradov
Department of Environmental Engineering, Faculty of Technical Sciences, University of Novi Sad, Novi Sad, Serbia
Heike Bradl
Department of Environmental Engineering, University of Applied Sciences Trier, Birkenfeld, Germany
Lea Kukoč-Modun
Department of Analytical Chemistry, Faculty of Chemistry and Technology, University of Split, Split, Croatia
Sanja Ćavar Zeljković
Centre of the Region Haná for Biotechnological and Agricultural Research, Department of Genetic Resources for Vegetables, Medicinal and Special Plants, Crop Research Institute, Šlechtitelů 29, Olomouc, Czech Republic
Centre of Region Haná for Biotechnological and Agricultural Research, Czech Advanced Technology and Research Institute, Palacky University, Šlechtitelů 27, Olomouc, Czech Republic
Igor Jerković
Department of Organic Chemisty, Faculty of Chemistry and Technology, University of Split, Split,Croatia
Roderick W. Bates
Division of Chemistry and Biological Chemistry, School of Physical and Mathematical Sciences, Nanyang Technlogical University, Singapore, Singapore
Safija Herenda
Department of Chemistry, Faculty of Science, University of Sarajevo, Sarajevo, Bosnia and Herzegovina
Muna Abu-Dalo
Department of Chemistry, Jordan University of Science and Technology, Irbid, Jordan

The journal is published semiannual, and full text version of the papers published are available free of cost at http://www.pmf.unsa.ba/hemija/glasnik.

Bulletin of the Chemists and Technologists has been licensed for indexing in:

Emerging Sources Citation Index (Web of Science, Clarivate Analytics)

CAPlus (Chemical Abstracts Plus):

Academic Search Complete (EBSCO)

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

2021
Issue 57

Print ISSN: 0367-4444
Online ISSN: 2232-7266

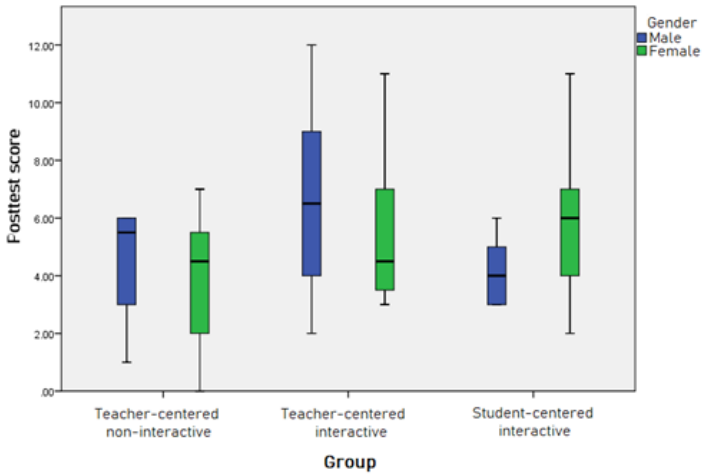
CONTENT

Editorial I

ORIGINAL SCIENTIFIC ARTICLES

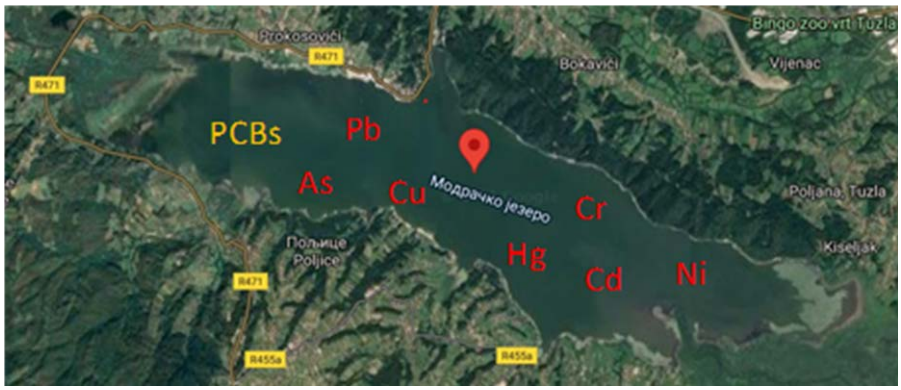
Seeking the right balance between three teaching approaches: a quasi-experimental study in the context of learning about thermal phenomena 1-8

Kolenda Darko
Vidak Andrej



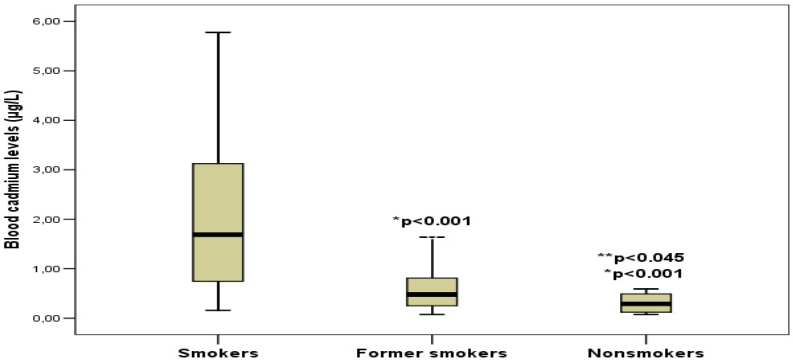
Occurrence of the heavy metals and PCBs in Accumulation Lake Modrac 9-16

Marjanovic Aleksandra
Djedjibegovic Jasmina
Omeragić Elma
Sober Miroslav



Smoking effect on the cadmium and zinc concentration in smokers and nonsmokers 17-26

Pepić Esad
Šečić Damir
Lepara Orhan
Kapić-Pleho Amna
Kurtagić-Pepić Emina
Mušanović Jasmin
Metović Azra
Džubur Alen
Naser Nabil
Panjeta Mirsad
Alić-Džubur Amela
Voljevica Alma



In memoriam – prof. dr. Mustafa Memić 27

Instructions for authors 29

Sponsors 37

Editorial

Potential drugs for oral treatment of COVID-19

A substantial number of patients with COVID-19, mostly older and those with preexisting chronic diseases need hospitalization, because of clinical progression to severe disease. Therefore, despite several vaccines in use, antiviral therapies that reduce the risk of COVID-19 progression are needed. Oral administration of such therapies would be ideally for the patients, who would easily use it by themselves. So far only one combination of antiviral drugs was approved for emergency use. As reported by manufacturer, combination of nirmatrelvir and low dose of ritonavir is an investigational SARS-CoV-2 protease inhibitor antiviral therapy, for oral administration. Nirmatrelvir is designed to inhibit replication of coronavirus, and low dose of ritonavir, helps maintaining adequate concentration of nirmatrelvir, by slowing down it's metabolism and breakdown. It is not authorized for the pre-exposure or post-exposure prevention of COVID-19 or for treatment in those requiring hospitalization due to severe or critical COVID-19. The U.S. Food and Drug Administration issued an emergency use authorization for this combination, which is different than an approval, in a sense that Agency determined that there is a reasonable believe that this combination can be effective in treatment of mild to moderate COVID-19 in authorized patients (adults and pediatric patients (12 years and older) with positive results of direct SARS-CoV-2 testing and who are at the risk for progression to severe COVID-19, including hospitalization or death), and that potential benefits outweigh the known and potential risks of the product. It is not authorized for use for longer than five consecutive days. So far there are no adequate, approved and available alternatives for the treatment of COVID-19, although some results of the phase 3 of clinical trial published on 16th December 2021, showed that oral molnupiravir (small-molecule ribonucleoside prodrug of n-hydroxycytidine) was found to be effective (significant reduction of hospitalization or death) for the early treatment of COVID-19 (within five days after the onset of signs and symptoms) without evident safety concerns, in the population of nonhospitalized, unvaccinated adults, who were at risk for the progression to severe disease.

Editors

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

2021

57

Print ISSN: 0367-4444
Online ISSN: 2232-7266

Original Scientific Article
DOI: https://doi.org/10.35666/2232-7266.2021.57.01
UDC: 542-053.5:37.018.2

1-8

Seeking the right balance between three teaching approaches: a quasi-experimental study in the context of learning about thermal phenomena

Kolenda, D.[a], Vidak, A.[b*]

[a]*Catholic School Center "Petar Barbarić" - Gymnasium Travnik, Bosnia and Herzegovina*
[b]*Faculty of Chemical Engineering and Technology, University of Zagreb, Croatia*

Article info
Received: 20/04/2021
Accepted: 30/09/2021

Keywords:
Work-Energy Principle
Rolling Motion
Gender Differences
Visualization

***Corresponding author:**
Andrej Vidak
E-mail: avdiak@fkit.hr
Phone: +38514597106

Abstract: In this study, we investigated which teaching approach may be optimal to facilitate learning about thermal phenomena in primary school. Concretely, we conducted a pretest-posttest quasi-experiment that included 45 eighth-grade students divided into three groups. In the first group (a non-interactive teacher-centered approach), the teacher gave an experiment-based lecture on converting thermal energy into mechanical work. In the second group (a teacher-centered interactive approach), the teacher gave the same experiments-based lecture, but interacted much more with the students and encouraged them to think about the demonstrations. Finally, in the third group, the student-centered interactive approach was applied. The results of the ANCOVA showed that the three teaching approaches were equally effective in developing students' understanding of thermal phenomena. However, closer analyses showed that students who learned from the teacher-centered interactive approach significantly outperformed their peers when it came to understanding basic thermal concepts approach, students worked in small groups to conduct the same experiments and "discover" the same relationships that the teacher had introduced in the previous one.

INTRODUCTION

Understanding thermal phenomena and thermodynamics is an important aspect of scientific literacy and this topic has found its place in science curricula at all educational levels. It is well known that many students have difficulty, understanding the basic concepts of thermal phenomena and thermodynamics (Vidak, Odžak & Mešić, 2019, Erickson, 1979, Carlton, 2000, Clough & Driver, 1985, Tiberghien's, Chu, Treagust, Yeo and Zadnik, Lewis (1996). In order to effectively overcome students' misconceptions, it is necessary to make additional efforts in the processes of conceptualization and conceptual change and therefore it is of great importance to design teaching lectures that will facilitate a better understanding of these phenomena. The literature shows that there is no consensus on which teaching approach is most useful in all physical contexts, so it is very important to determine the best approach for different contexts. One of the ways we can improve traditional teaching (a non-interactive teacher-centered approach) is to conduct experiments. In this approach, we can direct students closer to the theory and motivate them in studying physics phenomena. When performing the experiment, we can use the predict-observe-explain technique and predict-explain-observe-explain technique to foster conceptual understanding. The predict-observe-explain technique implies that students predict what will happen before performing the experiment, then observe what will happen and finally the students together with the teacher through a discussion come to an explanation of the observed phenomenon.The predict-explain-observe-explain technique consists of one additional element that is reflected in students explaining why they think what they predicted will happen even before the experiment is performed. Many studies have shown that research-based teaching (a teacher-centered interactive approach) can help students to increase their level of

conceptual understanding and gain a positive attitude toward science (Akar, 2005; Coulson, 2002; Tinnin, 2001). In the research of conceptual abilities using the CSEM test, students taught with the investigative teaching achieved higher results on the posttest i.e. students of four different lecturers achieved a score between 63% and 74%, while the average for their level of education was 47% (Etkina i Van Heuvelen, 2007). Furthermore, a comparative study conducted in 2004 showed that students taught by the investigative teaching method achieved 15% higher results than students taught by a non-interactive teacher-centered approach (73% of correct answers compared to 58% of correct answers) (Etkina i Van Heuvelen, 2007). In general, when using any teaching method it is important to consider the influence of cognitive load. Teachers often place high demands on working memory resources resulting in poor ability of students to learn the given materials. The working memory resources needed to learn a particular material (Sweller and Chandler, 1994) or to perform a particular task (Sweller et al., 1998) represent a cognitive load. The total cognitive load is equal to the sum of the intrinsic, relevant, and irrelevant cognitive load (Pass, Tuovinen, Tabers & Van Gerven, 2003). The relevant cognitive load leads to the adoption of new knowledge and automation of existing ones, as well as to the expansion of existing knowledge structures. By optimizing the intrinsic and minimizing the irrelevant cognitive load, the conditions for maximum student productivity are achieved. To achieve this goal (Van Merrienboer & Sweller, 2005) stated that it is useful to divide lectures into smaller “pieces” and provide explicit clues as well as to use external visualizations and analogies. Hardiman, Pollastek & Ewil (1986) and Brown & Campione (1994) stated that students who learn through investigative teaching with minimal feedback often become lost and frustrated, and their confusion can lead to misconceptions. Moreno (2004) concludes that there is a growing number of research showing that students learn with more understanding when investigative teaching is a more teacher-centered approach. In our study, we used all the scientific recommendations to balance the cognitive load of students and direct their attention to the optimal understanding of given thermodynamic phenomena.

Aim of the present study

The aim of our study is to investigate students’ conceptual understanding of the transformation of the inner energy into work, as well as the effects of different teaching approaches (teacher-centered interactive approach and student-centered interactive approach) compared to a non-interactive teaching teacher-centered approach. The results of methodological research in Bosnia and Herzegovina indicate that the interactive instruction approach is not sufficiently used (Suzić et al., 2009). The significance of this research is reflected in the fact that we will compare the achievement of a non-interactive teacher-centered teaching with two approaches of interactive teaching in developing a conceptual understanding of the transformation of internal energy into work and influence of individual teaching approaches to the aimed area. The results of this research can serve to improve the quality of interactive teaching implementation in the field of heat phenomena.

METHODOLOGY

Research design

In order to investigate the effectiveness of different teaching approaches, a quasi-experimental research was conducted as part of the regular classes determined by the curriculum. The total sample of students consists of three classes. One week before the start of the treatment a pretest was conducted. The pretest consisted of ten conceptual multiple choice questions that reflected the students’ misconceptions about the material relevant to the successful learning of the lecture “Turning internal energy into work". Seven days after the treatment, a posttest was conducted in which we measured the conceptual understanding of converting internal energy into work (closed-relevant conceptual questions) and elemental conceptual questions relevant to understanding the conversion of internal energy into work (broadly-relevant conceptual questions). To conduct pretest and posttest we allocated 25 and 35 minutes.

Participants

This study included 45 eighth-grade primary school students from the Primary School "Nova Bila" in Nova Bila, Bosnia and Herzegovina, of whom 20 were male and 25 female. The curriculum of the course includes that students learn about internal energy and heat in a two-hour lecture that is situated in the second semester of the school year. The total sample of students consists of three classes. One class received non-interactive teacher-centered treatment, while the remaining two received two different versions of the interactive teaching approach.

Curriculum and teaching treatment

In our research, quasi-experimental study was conducted as part of the regular class determined by the curriculum. Students in all three eighth grade classes were in their natural environment. When creating lectures, special accent was put on the content and conceptual questions. The questions in the introductory part of the lecture, experiments and the accompanying explanations, and the final part of the lecture were synchronized in all three groups. The questions from the pretest and posttest were not directly addressed during the treatment, but a theoretical basis for understanding these questions through the contents of the main part of the lecture was provided. It is well known that teacher-centered non-interactive teaching approach is characterized by one-way communication. Accordingly, the department taught in the teacher-centered non-interactive approach was characterized by one-way communication. The researcher solely performed a repetition of the relevant material, as well as the implementation of the main and final part of the lecture. In the class taught by teacher-centered interactive approach teaching students were interactively involved in the teaching process. The repetition of relevant material was conducted through conversation with students, the experiments were performed using the predict-explain-observe-explain

technique and the final part of the lecture was conducted through the questions which the researcher posted to the students. The third class was divided into four groups. The groups were composed of students of different abilities, according to the results of the pretest. Students conducted part of the lecture in independent activities, and the other part interactively with the researcher. The introductory part of the lecture was implemented through a conversation, after which the students were focused on group work. The group work has been conducted using the pre-created worksheets and experimental kit according to which students were guided step by step to the final explanation. After the students come to the final explanation of the outcome of the physical experiment through group work, the researcher explains it once again through a conversation with all the students. The final part of the lecture was conducted through a conversation between the researcher and the students.

RESULTS AND DISCUSSION

a) Analysis of the pretest scores

Within the group taught by teacher-centered non-interactive approach (TNI), there were 12 participants (Male=4, Female=8), 18 participants were taught by a teacher-centered interactive approach (TI) (Male=10, Female=8) and 15 (Male=6, Female=9) were student-centered interactive (SI). The pretest results show that the mean value of the TNI-group, TI-group and SI-group was 2.83 (1.46), 3.67 (1.84) and 3.40 (1.50), respectively, while the maximum score that students could achieve in the pretest was 10. The minimum score achieved in the TNI-group was 1, while maximum was 5; minimum score achieved in TI-group was 0, while maximum was 6; minimum score achieved in SI-group was 0, while maximum was 5. Table 1 shows the summarized data. The conceptual questions in the pretest reflect the content of the chapter "Internal energy and heat" in the regular curriculum. According to the results, students have a lot of difficulties in understanding thermodynamic concepts. According to the data in Table 2, we can conclude that students' scores on many questions are below 50 % for each group. Scores on Q1, Q2, Q5 and Q9 are very low for each group. The questions Q1, Q2, and Q5 relate to heat transfer from one object to another, while question Q9 relates to the particle movement in a closed container. The cause of the low scores on questions Q1 and Q5 can be attributed to the fact that students relied on their sensory experiences. Accordingly, students for objects that feel colder claim to be at a lower temperature and conclude that the insulator between two objects act as a heat source. Numerous students answered the question Q1 claiming that the metal would have the lowest temperature (TNI=83.3%, TI=77.8% and SI=86.7%). Question Q2 identified eventual misconception within the physics language and terminology, which is closely related to the heat transfer. To this question, most students replied that coldness is transferred from water to eggs (TNI=66.7%, TI=77.8% and SI=86.7%). As mentioned earlier, research by Chu, Treagust, Yeo and Zadnik (2012) found that students observe a sweater as a heat source rather than a thermal insulator, as confirmed by our research. It is interesting to emphasize that the question Q6 identified an identical misconception in a different context and that groups TI and SI were more successful with this question. Finally, all groups were equally unsuccessful on question Q9. Numerous students chose answer D (TNI=41.2%, TI=50% and SI=40%) as the correct answer i.e. the particle trajectory is circular.

Table 1. The pretest achievements

	Number of participants	Male	Female	Mean value (standard deviation)	Minimum value	Maximum value
TNI-group	12	4	8	2.83 (1.46)	1	4
TI-group	18	10	8	3.67 (1.84)	0	6
SI-group	15	6	9	3.40 (1.50)	0	5

Table 2. Summary of the individual groups scores on the pretest

	Q1	Q2	Q3	Q4	Q5	Q6	Q7	Q8	Q9	Q10
TNI-group	16.7%	16.67%	33.3%	83.3%	16.7%	16.7%	16.7%	33.3%	16.7%	33.3%
TI-group	11.1%	16.67%	72.2%	72.2%	16.7%	33.3%	55.6%	66.7%	11.1%	11.1%
SI-group	6.7%	6.7%	66.7%	73.3%	13.3%	33.3%	20.0%	73.3%	13.3%	40.0%

Between-group differences are presented in the following two figures. The x-axis shows the percentage of students who achieved the score presented on the y-axis.
Figure 1 shows between-group differences on the pretest for the teacher-centered non-interactive group and the teacher-centered interactive group.

We can notice the significant deviation in the number of students who have score 4 in different groups. Furthermore, 11.1% of students have score 0 in the teacher-centered interactive group while in the teacher-centered non-interactive group there were no such students.

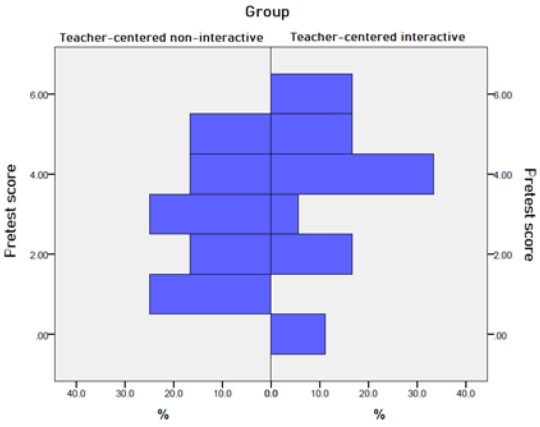


Figure 1. Between-group differences for teacher-centered non-interactive and teacher-centered interactive groups. The maximum score was 10.

If we compare the teacher-centered non-interactive and the student-centered interactive group (Figure 2), we can notice a deviation in some categories for the students from the SI group. We can see from the Figure 2 that 6.7% of students achieved a score 0 from the SI group, while there were no students with such score in the TNI group.

Furthermore, 25% of the students from the TNI group achieved score 1 on the pretest, while in the SI group the percentage was 0. Score 2, 4 and 5 on the pretest were achieved by 16.7% of the students from the TNI group while that percentage in the SI group was 26.7%, 33.3%, and 26.7%, respectively. If we consider the mean value, we can notice that there is no significant variation between groups and we cannot emphasize an overlap based on the test scores.

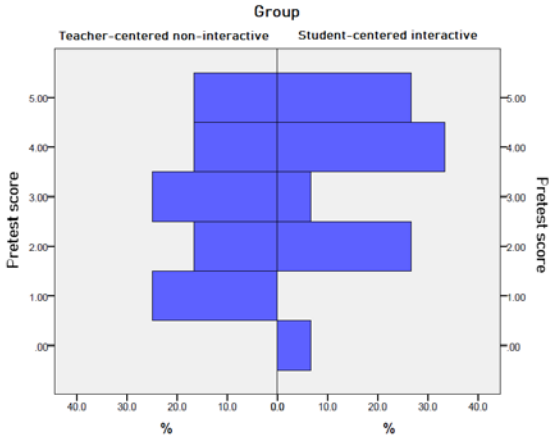


Figure 2. Between-group differences for teacher-centered non-interactive and student-centered interactive groups on the pretest. The maximum score was 10.

For the homogeneity of the variance on the pretest, we conducted Levene's test where the significance value was p=.728 (p>0.05). Accordingly, we can conclude that there is no significant variation between the group's variance achieved on the pretest.
We used the Kolmogorov-Smirnov test to check normality of the distribution. For the TNI-group, TI-group and SI-group significances were p=0.200, p=0.008 and p=0.010, respectively. Since the significance for the TI group is p=0.008 (p<0.05) we can conclude that the distribution varies from the normal one. We used Kruskal-Wallis's test whose significance was p=.309, from which we can conclude that there is no statistical significance between groups on the pretest. Table 3 shows between-group differences on the pretest scores.

Table 3. Between-group differences on the pretest scores

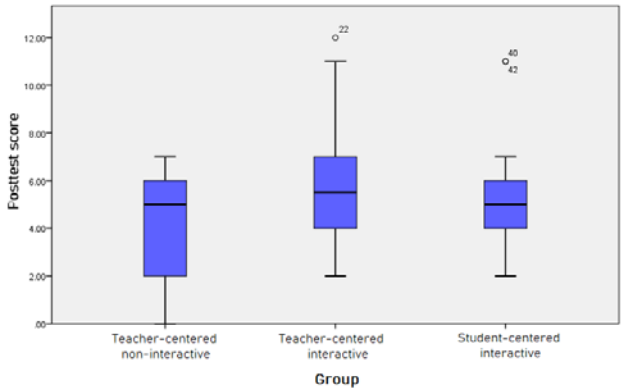
(I) Group	(J) Group	Significance
Teacher-centered non-interactive	Teacher-centered interactive	.370
Teacher-centered interactive	Student-centered interactive	.888
Student-centered interactive	Teacher-centered non-interactive	.649

b) Analysis of the posttest scores

The posttest consisted of fourteen questions, three of which had a multiple sub-questions. All questions were conceptual questions divided into two main groups: closely relevant and broadly relevant questions. Broadly-relevant questions measured the knowledge needed to conceptually understand the target lecture, while closely-relevant questions were closely related to the conversion of internal energy into work. It is important to know that none of the questions from the pretest were on the posttest.

Boxplot diagram in the Figure 3 shows that the median in the teacher-directed interactive group is slightly higher compared to medians in the other two groups (by 0.5). The median for the teacher-centered non-interactive and student-centered interactive group is equal and amounts to 5.00. Score intervals for teacher-centered non-interactive, teacher-centered interactive and student-centered interactive groups are 7.00 (min=.00. max=7.00), 10.00 (min=2.00, max=12.00) and 9.00 (min=2.00, max=11.00), respectively.

Figure 3 Score distribution on the posttest



Furthermore, Figure 4 shows the boxplot diagram for different genders. We can notice that the median is higher for boys in the first two groups, while the median for girls is higher in the student-centered interactive group.

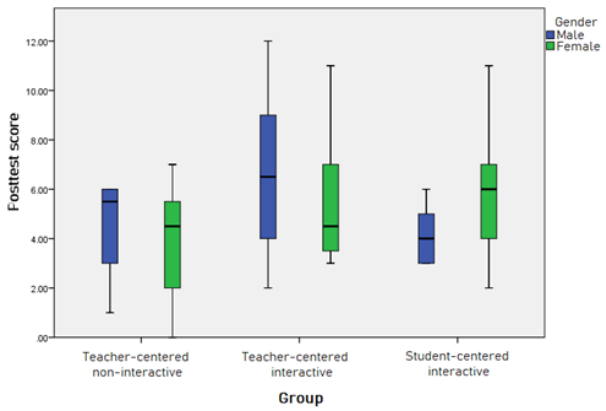


Figure 4. Score distribution according to gender

Table 4 shows the total score S, mean value (MV), standard deviation (SD) and variance (V) in each group, for closely relevant (CR) and broadly relevant (BR) questions, as well as the total value for each question on the posttest. We can immediately notice that the mean value is the highest for the teacher-centered interactive group and amounts to 6.12, for the student-centered interactive group the mean value is 5.40, while for teacher-centered non-interactive it is the lowest and equals to 4.08. Furthermore, we can observe the same order of groups when it comes to the mean value within broadly relevant questions. However, the mean value for closely-relevant questions for the student-centered interactive group is 2.73, for the teacher-directed group is 2.56, and for the teacher-centered non-interactive group is 2.08. We can conclude that the mean value of the scores for the experimental groups for each of the categories is higher than the mean value of scores for the teacher-centered non-interactive group.

Table 4. Scores on the posttest

	Teacher-centered non-interactive group		
	CR	BR	S
MV	2.08	2.00	4.08
SD	1.16	1.60	2.27
V	1.36	2.54	5.17
	Teacher-centered interactive group		
	CR	BR	S
MV	2.56	3.56	6.12
SD	2.38	1.29	2.89
V	5.67	1.67	8.34
	Student-centered interactive group		
	CR	BR	S
MV	2.73	2.67	5.40
SD	2.05	1.05	2.64
V	4.21	1.10	6.97

We can gain a deeper insight into student's progress by considering the score difference on the pretest and the posttest. Since there were ten questions on the pretest, while there were fourteen on the posttest, we perform the scaling of the results by multiplying the pretest score with the factor of 1.4. After that, we performed the following coding: students who scored 0 to 2.8 were put in the low-ability group (L), students who scored 2.8 to 5.6 were put in the medium-ability group (M), and the students who scored 5.6 to 8.4 were put in the high-ability group (H). After scaling and placing the students

in their respective ability groups, we measured the score difference on the pretest and posttest, which is summarized in the following table 5.

Table 5. Mean values difference regard the ability groups

Ability level	Teacher-centered non-interactive
Low	1.84
Medium	-0.76
High	-2.00
	Teacher-centered interactive
Low	4.12
Medium	0.31
High	-0.87
	Student-centered interactive
Low	3.56
Medium	0.13
High	-2.25

We can notice that the average difference of the scores has a tendency to decrease for all groups. Based on this fact, the students from the low-ability group within the teacher-centered interactive group made the greatest progress, followed by students from the low-ability group within the student-centered interactive group, and finally the students from the low-ability group within the teacher-centered non-interactive one. We need to consider this fact with special care. Better insight into group differences could be achieved by using the same pretest and posttest to obtain highly valid considerations. The number of students is too small and there is a danger of the statistical inference (for example, statistically speaking, there is a higher probability for progress of a students who achieved a lower score on the pretest).

All previous considerations were based on the mean value of the scores achieved on the pretest and posttest. Now, we use inferential statistic to determine if there is statistically significant difference between groups. For this purpose, the analysis of covariance is used. Score on the pretest is used as a covariate, while the dependent variables are total pretest scores, followed by the scores on the broadly-relevant questions and, finally, the scores achieved on the closely-relevant questions. To better understand how the covariance (scores on the pretest) adjusts the original mean value and the standard deviation, we will refer to the tables 6 and 7.

Levene's coefficient for these data is .680, from which it is possible to conclude that variances of the score achieved in the posttest are homogeneous. By analyzing the between-group differences, with the posttest as a dependent variable, we determine that the difference between groups are not so statistically significant [F(2,41)=1.719, p=.192].

When we choose the posttest scores on the closely-relevant questions as a dependent variable, Levene's coefficient is .042, which could mean that the results are less reliable. When it comes to the analysis of the covariance, we conclude that there is no statistically significant difference between the presented groups [F(2,41)=.351, p=.706].

Table 6. Mean values on the posttest, closely-relevant and broadly-relevant questions. Standard deviations are given in parentheses.

Mean value	Posttest	Closely-relevant questions	Broadly-relevant questions
Teacher-centered non-interactive	4.083 (2.275)	2.083 (1.164)	2.000 (1.595)
Teacher-centered interactive	6.111 (2.888)	2.556 (2.382)	3.556 (1.294)
Student-centered interactive	5.400 (2.640)	2.733 (2.052)	2.667 (1.047)

Table 7. Adjusted mean values on the posttest, closely-relevant and broadly-relevant questions. Standard deviations are given in parentheses.

Adjusted mean value	Posttest	Closely-relevant questions	Broadly-relevant questions
Teacher-centered non-interactive	4.173 (.783)	2.079 (.597)	2.094 (.377)
Teacher-centered interactive	6.058 (.635)	2.558 (.484)	3.500 (.306)
Student-centered interactive	5.392 (.690)	2.734 (.526)	2.659 (.333)

Finally, we select broadly-relevant questions as the dependent variable. The Levene's coefficient is equal to .061, which is not statistically significant and we can conclude that the variances are homogeneous. Here we obtain a statistically significant difference between the groups [F(2,41)=4.343, p=.019]. Since we find statistically significant differences between some groups, we need to conduct post hoc testing. Using the Bonferroni test, a statistically significant difference was found between the non-interactive teacher-centered and the teacher-directed interactive group (p=0.20).

Therefore, a statistically significant difference is determined between the teacher-centered non-interactive and the teacher-centered interactive groups in the domain of the broadly-relevant questions. In terms of achievement on the posttest, closely-relevant, and broadly-relevant questions, there are no statistically significant differences among groups, with regard to gender.

By analyzing the pretest, we perceive a poor understanding of the basic concepts of thermodynamics. During the treatment, the lecture "Converting internal energy into work" was processed in a way that the

curriculum was simplified and after that, the experiment was conducted with students. On the theoretical side, we can say that students studied by manipulating existing knowledge to gain more complex knowledge. Since most students did not have a well-developed knowledge to understand the complex material (Converting internal energy into work), it was expected that they would not sufficiently develop the coherent structures of knowledge. Instead, students adopted more fundamental knowledge during the treatment. Statistically speaking, the teacher-centered interactive group achieved a statistically significant difference when it comes to the basic thermodynamic concepts, compared to students taught in a non-interactive teacher-centered approach.

During the student-centered interactive physics teaching, students conduct most of the activities independently, which could be the reason for the poor understanding of the lecture "Converting internal energy into work". Students tried to gain some insight into more complex aspects by using the knowledge they already had, but as they did not have a sufficiently developed knowledge, it was expected that they did not sufficiently understand the new lecture. The student-centered interactive teaching method would probably be more effective if we use more time to implement it, which is in line with the fact that this type of teaching takes more time than the teacher-centered non-interactive teaching method.

CONCLUSION

The goal of this study was to compare the effectiveness of two types of interactive teaching compared to traditional teaching. To accomplish this goal, we conducted research in which we measured students' conceptual understanding of basic thermodynamic concepts in the pretest, and conceptual understanding of thermodynamic concepts (broadly relevant questions) and converting internal energy into work (closed relevant questions) on the posttest. Three eighth-grade classes participated in the study, with one department being taught in the teacher-centered non-interactive, the other as teacher-centered interactive, and the third through student-centered interactive approach.

It is very important for different groups of students in different contexts, to investigate what is the relationship of time spent on what will lead to the best learning of selected topics. In this study, we examined this relationship for eighth grade' students from Bosnia and Herzegovina in the context of learning about converting internal energy into work. The obtained results show that the least effective was the teacher-centered non-interactive approach. These results can be related to the fact that in this approach students are not encouraged to implement higher thought processes, i.e. time is not used effectively to facilitate learning. On the other hand, a student-centered interactive approach has many advantages to foster higher thought processes, but many students have difficulty managing the entire learning process independently, which can lead to cognitive overload. The teacher-centered interactive approach seems optimal for the age of students who are just beginning to learn physics - this approach seems to offer optimal balance through timely class discussions.

REFERENCES

Akar, E. (2005). Effectiveness of 5E learning cycle model on students' understanding of acid-base concepts. PhD thesis, METU.

Brown, A. L. and Campione, J. C. (1994). Guided discovery in a community of learners. The MIT Press.

Carlton, K. (2000). Teaching about heat and temperature. Physics Education, 35(2):101.

Chu, H.-E., Treagust, D. F., Yeo, S., and Zadnik, M. (2012). Evaluation of students' understanding of thermal concepts in everyday contexts. International Journal of Science Education, 34(10):1509-1534.

Clough, E. E. and Driver, R. (1985). Secondary students' conceptions of the conduction of heat: Bringing together scienti_c and personal views. Physics Education, 20(4):176-82.

Coulson, D. (2002). Bscs science: An inquiry approach-2002 evaluation findings. Arnold, MD: PS International.

Erceg, N., Aviani, I., Me_si_c, V., Glun_ci_c, M., and _Zauhar, G. (2016). Development of the kinetic molecular theory of gases concept inventory: Preliminary results on university students' misconceptions. Physical Review Physics Education Research, 12(2):020139.

Erickson, G. and Tiberghien, A. (1985). Heat and temperature. Children's ideas in science, pages 52-84.

Etkina, E., & Van Heuvelen, A. (2007). Investigative science learning environment–A science process approach to learning physics. Research-based reform of university physics, 1(1), 1-48.

Hardiman, P. T., Pollatsek, A., and Well, A. D. (1986). Learning to understand the balance beam. Cognition and Instruction, 3(1):63-86.

Lewis, E. L. (1996). Conceptual change among middle school students studying elementary thermodynamics. Journal of Science Education and Technology, 5(1):3-31.

Moreno, R. (2004). Decreasing cognitive load for novice students: Effects of explanatory versus corrective feedback in discovery-based multimedia. Instructional science, 32(1-2):99-113.

Paas, F., Tuovinen, J. E., Tabbers, H., and Van Gerven, P. W. (2003). Cognitive load measurement as a means to advance cognitive load theory. Educational psychologist, 38(1):63-71.

Suzić, N., Živković, S., Alić, A., Skelić, D., Rukavina, D., Alibegović Goro, E., Džumhur, Ž., Šahinović Batista, S., Milinković Rosić, I., Mešić, V., and Ibraković, A. (2009). Sekundarna analiza TIMSS 2007 u Bosni i Hercegovini. Agencija za predškolsko, osnovno i srednje obrazovanje.

Sweller, J. and Chandler, P. (1994). Why some material is difficult to learn. Cognition and instruction, 12(3):185-233.

Sweller, J., Van Merrienboer, J. J., and Paas, F. G. (1998). Cognitive architecture and instructional design. Educational psychology review, 10(3):251-296.

Tiberghien, A. (1980). Modes and conditions of learning. an example: the learning of some aspects of the concept of heat. Cognitive development research in science and mathematics, pages 288-309.

Tinnin, R. K. (2001). The effectiveness of a long-term professional development program on teachers' self-efficacy, attitudes, skills, and knowledge using a thematic learning approach.

Van Merrienboer, J. J., & Sweller, J. (2005). Cognitive load theory and complex learning: Recent developments and future directions. Educational psychology review, 17(2), 147-177.

Vidak, A., Odžak, S., & Mešić, V. (2019). Teaching about thermal expansion: investigating the effectiveness of a cognitive bridging approach. Research in Science & Technological Education, 37(3), 324-345.

Summary/Sažetak

U ovoj smo studiji istražili koji nastavni pristup može biti optimalan za olakšavanje učenja o toplinskim pojavama u osnovnoj školi. Konkretno, proveli smo kvazi eksperimentalno istraživanje koje je obuhvatilo 45 učenika osmih razreda podijeljenih u tri skupine. U prvoj skupini (ne-interaktivni pristup usmjeren na nastavnika), nastavnik je održao predavanje sa eksperimentima o pretvaranju toplinske energije u mehanički rad. U drugoj skupini (interaktivni pristup usmjeren na nastavnika) nastavnik je održao isto predavanje temeljeno na eksperimentima, ali je mnogo više komunicirao sa učenicima i potaknuo ih da razmišljaju o predstavljanim eksperimentima. Konačno, u trećoj skupini (interaktivni pristup usmjeren na učenika) učenici su radili u malim skupinama kako bi izveli jednake eksperimente i „otkrili“ odnose koje je nastavnik uveo u prethodne dvije skupine. Rezultati ANCOVA-e pokazali su da su jednaku učinkovitost sva tri nastavna pristupa u razvijanju razumijevanja učenika o toplinskim pojavama. Međutim, detaljnije analize pokazale su da učenici koji su učili korištenjem interaktivnog pristupa usmjerenog na nastavnika značajno nadmašuju svoje vršnjake kada se promatra razumijevanje osnovnih toplinskih pojmova.

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

2021

57

Print ISSN: 0367-4444
Online ISSN: 2232-7266

Original Scientific Article
DOI: https://doi.org/10.35666/2232-7266.2021.57.02
UDC: 543[628.312.2:549.28](497.-18 Modrac)

9-16

Occurrence of the heavy metals and PCBs in Accumulation Lake Modrac

Marjanovic, A.*, Djedjibegovic, J., Omeragić, E., Sober, M.

Faculty of Pharmacy, University of Sarajevo, Zmaja od Bosne 8, 71 000 Sarajevo, BiH

Article info
Received: 14/06/2021
Accepted: 13/12/2021

Keywords:
Accumulation
Lake Modrac
PCBs
heavy metals

***Corresponding author:**
Aleksandra Marjanović
E-mail: aca1902@gmail.com
Phone: +387 61 709 562

Abstract: The accumulation Lake Modrac is a particularly important source of drinking water for inhabitants of the Tuzla region and few local settlements. The most significant point sources of organic contaminants in the accumulation Lake Modrac are waste water from households and industry. In this area, most of the settlements have neither sewage systems nor facilities for waste water treatment. Other potential point sources of pollutants are industrial plants. The most prominent are coal mines (Banovići and Đurđevik), metal and wood industry, plant for plastic production, and oil and oil derivatives warehouse. Few previously conducted surveys in the region showed the presence of the persistent organic pollutants and heavy metals in large extent. The objective of this study was to conduct a water quality survey targeting selected inorganic (Cd, Cr, Cu, Hg, Ni, Pb and As) and organic pollutants in the accumulation Lake Modrac in Bosnia and Herzegovina. The content of polychlorinated biphenyls (PCBs) determined with ELISA test, ranging from 3.23 to 6.19 µg/L (sum of 7PCBs).The most abundant metals (analyzed by graphite furnace AAS and mercury analyzer) at all five sampling locations were Pb (6.79-36.58 µg/L); Ni (5.81-10.43 µg/L) and Hg (1.08-6.10 µg/L).

INTRODUCTION

Accumulation Lake Modrac is the largest and most important multi-purpose water resource for water supply of large important industrial capacities in Bosnia and Herzegovina, as well as for public water supply of Tuzla and Lukavac. In addition, main tributaries of accumulation Lake Modrac, River Jala and River Spreča, are also main tributaries (River Spreča) of River Bosna, which is a transboundary river, entering Croatia (tributary to River Sava). Therefore, the Bosna River is a source of pollutants for the Danube, where it eventually drains. The water quality of the accumulation is threatened by the wastewater from communities and industrial facilities (the concentrated polluters) which discharges directly into the accumulation, most of them without any treatment. According to the Coordinating team for protection of accumulation Lake Modrac (Strategy, 2012), the most prominent point sources of pollutants are coal mines (Banovići and Đurđevik), metal and wood industry, plant for plastic production, oil, and oil derivatives warehouse.

All these facilites discharges mainly untreated wastewater into the Lake Modrac or its tributaries. The useful volume of accumulation is limited, primarily due to prolonged deposition of coal dust from coal mines in the basin reservoirs (Strategy, 2012).

The diffuse sources of pollution of the accumulation Lake Modrac with persistent organic pollutants (POPs) are chemical contamination of land used for agricultural production, unregulated landfill for municipal and industrial waste, forestry, transportation, atmospheric deposition of air pollutants, leaching from urban surfaces and industrial areas and others.

In the area of the Bosna River basin, there is a high number of landfills that often contain medical waste (beside municipal and industrial waste), which poses a significant risk and affects the water quality.

Uncontrolled landfills for municipal and other waste are very often the source of fires causing the emission of large quantities of toxic pollutants, including polychlorinated biphenyls (PCB), polychlorinated

dibenzodioxins (PCDD) and polychlorinated dibenzofurans (PCDF).

During the war (1992-1995) in Bosnia and Herzegovina, large amounts of POPs were generated and emitted into the environment because of partial or complete destruction of industrial facilities, military targets, infrastructure, explosions, and uncontrolled fires (Klánová, Kohoutek, Kostrhounová, *et al.*, 2007).

Persistent pollutants are suspected to be a potential problem in major rivers in Bosnia and Herzegovina. However, very little information exists on persistent pollutants contamination as very little analyses have been done (Harman, Grung, Djedjibegovic, *et al.*, 2013; Harman, Grung, Djedjibegovic, *et al.*, 2018).

Studies of environmental quality or water quality of Lake Modrac dated back after the war (latest data are from 1991) and are scarce. In 2016 investigation that was conducted by the Institute for chemical engineering Tuzla (as ordered by public water management company Spreča-Tuzla) showed worsening of all physicochemical parameters of water quality in Lake Modrac comparing to similar investigations done in 2015 (Institute for chemical engineering, 2016). According to legislation in Federation of Bosnia and Herzegovina (FBiH), Lake Modrac should be categorized as class II water, meaning that after physical treatment, chemical treatment, and disinfection this water can be used as drinking water, for aquaculture of Salmonids or for the recreative purposes (swimming or sport) (Directive, 1967).

Results of physicochemical analysis of surface water (JP Spreča dd, laboratory) conducted in 2017 showed that water from the Lake Modrac can be classified as class II, III, IV, or sometimes even non categorized (mouth of River Spreča into a Lake Modrac). Regarding to content of total suspended solids (TSS) it can be categorized as class IV (Reports JP Spreča, 2017). High content of TSS is usually due to contamination with wastewater, or it can point to diffuse sources (*i.e.*, agricultural activities).

Calculated chemical oxygen demand/biochemical oxygen demand (COD/BOD_5) ratio was 5.21-8.9 (usually is 1.5-3.0; in extreme cases >5) (Reports JP Spreča, 2017) and is obvious sign of pollution. Inversely, BOD_5/COD ratio calculated from the above presented data from JP Spreča Report is 0.11-0.19. BOD_5/COD ratio 0-0.5 can suggest presence of persistent organic compounds (*i.e.*, coal dust, lignins, tannins, cellulose, synthetic organic compounds) (Lee, Lee, Yu, *et al.*, 2016).

Heavy metals and POPs can pose a significant risk for the environment and human health. So far, great number of studies have linked exposure to POPs with diseases and abnormalities in wildlife species, including fish, birds and mammals. In humans POPs can cause variable adverse health effects, especially, reproductive, neurological, behavioral, immunological and endocrine (Li, Loganath, Chong, *et al., 2006;* Kumar, Kumar Sarma, Shubham, *et al.*, 2020).

Literature data suggest that all classes of POPs including PCBs and other pollutants like some heavy metals/metalloids, such as Cd, Pb, Hg and As can be classified as endocrine disrupting compounds (EDCs). For example, according to results of previously conducted studies, exposure to low levels of certain essential or trace metals (*i.e.*, Cu, Cr) can also be related to adverse effects on reproduction in humans (Ashrap, Meeker, Sánchez, *et al., 2020;* Street, Angelini, Bernasconi *et al., 2018;* Street, Angelini, Bernasconi, *et al.*, 2018; Nwadiuto Amadi, Nkeiruka Igweze, Ebere Orisakwe, 2017). Special problem is possible additive toxicity as result of exposure to a mixture of toxicants that can act on the same organs or tissues.

Bearing this in mind, the surveillance of specific pollutants, representing the level of anthropogenic impact in the water bodies, is necessary for chemical water body status classification. Among others, preserving and protecting the "'good status'' for all waters, surface and groundwater, is one of the main goals of Water Framework Directive (WFD) (EC, 2000). Survey and monitoring data are important basis for sound management of the water resources. Therefore, the objective of this study was to conduct a water quality survey targeted on selected inorganic (Cd, Cr, Cu, Hg, Ni, Pb and As) and organic pollutants (PCBs) in the accumulation Lake Modrac in Bosnia and Herzegovina.

EXPERIMENTAL

Sampling

Samples (N=5) analyzed in this work were taken from different locations at accumulation Lake Modrac in August 2018. Sampling sites are presented in Table 1.

Table 1. Sampling locations at accumulation Lake Modrac

Sample	Locations	Coordinates
M1	Mouth of River Turija	44°30'31.6"N 18°27'53.6"E
M2	Trnovčica	44°29'40.2"N 18°30'05.1"E
M3	Mouth of River Spreča	44°28'42.2"N 18°32'30.6"E
M4	Prokosovići	44°30'37.0"N 18°29'01.1"E
M5	Behind the dam of company "Spreča"	44°30'57.0"N 18°30'37.9"E

For the metal analysis, water samples (2 L) were taken in clean plastic bottles, stabilized by adding 2 ml of concentrated nitric acid (67-69 % HNO_3, Optima grade, Fisher Scientific, UK) and stored in refrigerator (4 ^{0}C) prior the analysis. All the samples were analyzed in triplicate.

For the analysis of PCBs water samples (1 L) were collected in glass containers, previously washed with methanol (extra pure, Fisher Scientific, UK), and dried in the oven (400 ^{0}C) for a couple of hours. Containers were fitted with the foil-lined screw caps. Samples were diluted with the equal volume of methanol to prevent adsorptive losses.

Materials

All used chemicals were of ultra-pure grade. The water (18.2 μScm^{-1}) used for the preparation of reagents and standards was obtained from a laboratory water purifying system (Arium 611, Sartorius Mechatronics, Goettingen, Germany).

All the glassware and utensils used for the metal analysis were washed with the ultra-pure water, after soaking in 10 % HNO_3 for 24 h
All the glassware and utensils used for the analysis of PCBs were washed with ultra-pure water, rinsed with methanol and dried (400 ^{0}C) in the oven for a couple of hours.

Sample preparation for heavy metal analysis
For the analysis of heavy metals/metalloids (Pb, Cd, total As, Cu, Cr, Ni), samples were digested in the microwave oven Ethos D 118 (Milestone, Sorisole, Italy) in accordance with standard method (BAS EN 13805:2015) previously described by Omeragić *et al.* (Omeragic, Radosevic, Causevic, *et al.*, 2021).

Analysis of selected heavy metals
Analysis of the selected heavy metals in water samples was performed at the laboratory of the Institute for Public Health of the Federation of Bosnia and Herzegovina. For the determination of Pb, Cd, total As, Cu, Cr and Ni graphite furnace atomic absorption spectrometry (GFAAS) analysis was performed using the atomic absorption spectrometer AA-7000F Dual Atomizer System (Shimadzu, Tokyo, Japan), equipped with self-reversal method (SR) background correction. For the removal of residues from the graphite tube during pyrolysis, argon flow was used. Analysis was done using the standard method for the determination of trace metals using GFAAS (BAS EN ISO 15586:2005).
The content of Hg was determined using AMA 254 advanced mercury analyzer (Leco Inc., USA) with the *in-house* method. Briefly, the sample was directly measured (ranged between 0.1 and 0.5 ml) into the precleaned sample boat and placed in the instrument. The Hg concentration was determined using the principle of thermal decomposition, amalgamation, and atomic absorption spectrometry at 254 nm wavelength. A standard solution of Hg (1000 mg/L±2 mg/L, Merck KGaA, Darmstadt, Germany) was diluted with HNO_3 to obtain working solutions for preparation of calibration curves (2.5–30.0 ng and 100–500 ng).

Table 2. Parameters of used analytical methods

Metal	Certified concentration (μg/L)	% RSD	Recovery (%)	LOQ (mg/L)
Cd	6.568	0.014	89.8	0.00002
Cu	22.76	0.004	97.7	0.0014
Pb	19.63	0.010	88.6	0.0011
As	60.45	0.001	96.9	0.0011
Ni	62.41	0.001	95.9	0.0019
Cr	20.40	0.000	87.4	0.0004
Hg	10.00	0.010	92.4	0.0004

* RSD-relative standard deviation; LOQ-limit of quantification

The blanks and standards were analyzed with the same methods as samples. Blanks did not contain detectable amounts of the analyzed metals.

In order to verify the validity of the measurements, standard reference materials, NIST SRM 1643f - Trace Elements in Water (for Cd, Cr, Cu, Ni, Pb and As) and NIST SRM 3133 - Mercury (Hg) Standard Solution were used. The recovery was calculated as the percentage of the certified concentration of a metal in the standard reference material determined during the analytical procedure. Analytical method parameters are presented in Table 2.

Analysis of PCBs
For the determination of PCBs in the analyzed samples Abraxis PCB kit was used. This kit is based on the principles of the enzyme linked immunosorbent assay (ELISA) and is intended to use for the determination of PCBs in water samples.
Samples were prepared for the analysis and analyzed according to a procedure provided by the manufacturer of the kit. In brief, after collecting, samples were diluted and filtered using the glass fiber syringe filter (0.45 μm). Aliquots (50 μL) of samples were transferred to a well of the microtiter plate and mixed with the enzyme conjugate solution (the horseradish peroxidase labeled PCB analog), antibody solution (rabbit anti-PCB), substrate (colour) solution (hydrogen peroxide and 3,3',5,5'-tetramethylbenzidine in organic base) and stop solution (0.5 % sulfuric acid) in consecutive steps following the provided protocol (Abraxis, 2018).
Results of testing (*i.e.*, content of PCBs) are expressed as Aroclor 1254 (μg/L).
As a part of the quality control the kit has included a control solution (approximately 5 μg/L of PCB) that is included in every run and handled in the same way as samples. For the determination of the concentration of PCBs in samples, standard solutions of PCBs (as Aroclor 1254) provided within the kit were used (0, 1, 5, 25, 100, 250 μg/L). Calibration curve is constructed by plotting the logit B/B_0 for each standard versus the natural logarithm of the corresponding PCB concentration.
Logit value is calculated using equation 1:

$$\text{Logit } B/B_0 = \log \frac{\frac{A}{A_0}}{1-\frac{A}{A_0}} \qquad (1)$$

where A_0 is average absorbance value for the negative control (standard concentration 0), and A is average absorbance value for the standard/sample.

RESULTS AND DISCUSSION

The content of metals determined in water samples (μg/L) is presented in Table 3. with side-by-side view of the maximum allowable concentrations (MAC) and target values (TV) for the specific metals proposed in the Council Directive 75/440/EEC concerning the quality required of the surface water intended for the abstraction of drinking water in the Member States (EC, 1975), Decision on characterization of surface and underground waters, reference conditions and parameters for the estimation of the water status and water monitoring in

FBiH (Decision, 2014), EU Directive 2013/39/EU on environmental quality standards in the field of water policy (EC, 2013) and Dutch environmental quality standards (EQS) for surface water and sediments (Warmer and van Dokkum, 2002).

Table 3. The concentration of metals (μg/L) in water samples and maximum allowable concentrations defined by FBiH legislation and EU directives

Sample	Metals (μg/L)						
	Pb	Cd	As	Cu	Cr	Ni	Hg
M1	36.58±0.214	0.283±0.014	1.386±0.023	3.117±0.141	1.255±0.001	6.920±0.017	1.075±0.035
M2	8.573±0.065	0.024±0.000	1.474±0.001	2.267±0.047	1.129±0.000	6.920±0.030	2.495±0.008
M3	6.790±0.157	0.031±0.001	1.892±0.003	3.202±0.004	0.859±0.059	10.43±0.001	6.098±0.003
M4	7.532±0.010	0.064±0.000	2.349±0.003	4.175±0.038	0.558±0.049	7.900±0.020	3.893±0.011
M5	18.16±0.000	0.066±0.000	3.518±0.003	2.724±0.016	0.453±0.000	5.813±0.020	5.113±0.018
Limits							
75/440/EEC (mg/L)	0.05	0.005	0.05	0.05	0.005	-	0.001
Decision FBiH MAC/EQS (μg/L)	7.20[a]	≤0.45-1.50	20.0	1.10-8.80[b]	10.0	20.0[a]	0.07
2013/39/EU (μg/L)	14.0	0.45-0.60				34	0.07
Dutch EQS MAC (μg/L)	220	2.0	32.0	3.80	84.0	6.30	1.20
Dutch EQS TV (μg/L)	5.30	0.40	1.30	1.10	2.40	4.10	0.07

Legend: [a]-non applicable MAC-EQS, annual average (AA) is applied; [b]-depending on the hardness of water

The content of all metals except Hg at each of five locations was lower than limits defined in the Council Directive 75/440/EEC. The concentration of Hg recorded at location 1 (1.075 μg/L) was slightly higher than the defined limit (1 μg/L), while on the other four locations concentrations were two to six times higher than the limit values (Table 3).

Beside Hg the most abundant metals at all locations were Pb and Ni. The highest content of Pb was found at locations 1 and 5 (36.58 and 18.16 μg/L, respectively), while the highest concentration of Ni was found in samples from the locations 3 and 4 (10.43 and 7.90 μg/L, respectively).

The concentration of Pb, As, Cu and Ni in all samples from all 5 locations were higher than target values defined in Dutch EQS (Table 3). Measured concentration of Cu (4.175 μg/L) in sample from the location 4 was higher than MAC defined in Dutch EQS (3.8 μg/L), as well as concentrations of Ni at locations 1-4 (>6.3 μg/L).

The concentration of Hg at all 5 locations was slightly higher than MAC defined in Directive 2008/105/EC and target value defined in Dutch EQS (0.07 μg/L) and two to five times higher than MAC defined in Dutch EQS (Table 3).

Concentration of Cd at all five locations was well below all aforementioned limits.

Comparing the obtained results to the legislation in FBiH, concentrations of As, Cd, Cu, Cr and Ni were bellow MAC or annual average, while concentration of Hg at all five locations was almost two orders of magnitude higher than MAC (0.07 μg/L). Concentration of Pb was slightly below annual average value (6.79 μg/L vs 7.2 μg/L) only at one location (M3).

Results of our measurement were also compared with the limits defined in Directive on classification of the surface water and water bodies categorization in Republika Srpska (RS) (Directive, 2001). Maximum permitted levels for inorganic substances (metals and metalloids) for the class II and III (equivalent to class II for the Lake Modrac) are presented in Table 4.

Table 4. Maximum permitted levels for inorganic substances (metals) for the water bodies class II and III

Metals (total)	class II (μg/L)	class III (μg/L)
As	10-20	20-40
Cd	0.05-1.0	1.0-2.0
Cr	5-15	15-30
Cu	5-15	15-50
Ni	0.05-1.0	1.0-2.0
Pb	0.1-0.5	0.5-2.0
Hg	0.1-0.2	0.2-0.5

Registered concentrations of As, Cr, Cu and Cd in all analyzed samples (Table 3) were bellow permitted limits defined in aforementioned Directive (Table 4), while the concentrations of Pb, Ni and Hg were slightly higher than limits in all analyzed samples.

There is almost no data on contamination of the Lake Modrac with heavy metals and organic pollutants published in the literature. Comparing the obtained results in this study with the study conducted in 2007 on the presence of heavy metals at accumulation Lake Modrac, average concentration of Pb and Cd measured in this study was two to ten times lower, while concentration of Cu was in average two to three times higher (Đonlagić, Odobašić, Bratovčić, 2007).

Overall average contents of Pb, Ni and Cd registered in this study at locations M1 and M5 (27.37 μg/L; 6.37 μg/L and 0.283 μg/L, respecitvelly), were lower than the results of the monitoring of Lake Modrac conducted in 2019 at the same locations (143.88 μg/L Pb; 8.28 μg/L Cd and 43.02 μg/L Ni) (Đozić and Alihodžić, 2019). Average concentrations of As (<0.05 μg/L) and Hg (<0.0001 μg/L) reported by this study were much lower than our results (2.45 μg/L As and 3.09 μg/L Hg).

In comparison to some other lake waters pollution in various countries, including Balkan region, our result are slightly higher than those reported for Lake Kalimanci (Macedonia) and clearly higher than in Skadar Lake (Montenegro), Kralkizi Reservoir, Dickle Reservoir, Batman Reservoir and Isikli Lake (Turkey) for all tested metals (Sibal and Espino, 2018).

Significant positive correlation was found for Pb and Cd concentrations (r=0.953, p=0.012), and significant negative correlation for As and Cr (r=-0.911, p=0.032).

Polichlorinated biphenyls (PCBs) in the samples from the accumulation Lake Modrac were determined using commercial ELISA kit. Results of measurement were expressed as content of Aroclor 1254 and sum of 7 indicator PCBs (congeners CB 28, CB 52, CB 101, CB 118, CB 138, CB 153 and CB 180) and presented in Table 5.

Table 5. Concentration of PCBs in analyzed samples from accumulation lake Modrac

Sample	Aroclor 1254 (μg/L)	Sum 7PCBs (μg/L)
M1	10.35±1.29	3.23±0.40
M2	19.84±7.47	6.19±2.33
M3	13.79±4.76	4.30±1.48
M4	17.94±5.80	5.59±1.81
M5	12.91±2.51	4.03±0.78

Aroclor 1254 is one of the several commercial mixtures of PCBs that were extensively used worldwide and still can be measured in environment. Concentrations of PCBs can be presented in few ways depending on the purpose of the analysis and method used. Commercial kits based on ELISA are recognized by Unites States Environmental Protection Agency (US EPA) for the screening purposes and often are used for the detection of presence of PCBs in the water or sediment.

Result of measurements are often expressed as Aroclor content in risk assessment studies, while in most ecotoxicology studies it is important to determine not only total concentration of PCBs, but also congener profile. In that case mostly low chlorinated and high chlorinated congeners (28, 52, 101, 118, 138, 153 and 180) or so called "7 Dutch" or 7PCBs are determined. For that reason, we have recalculated results of the measurements from this study, that were originally expressed as content of Aroclor 1254, to the content of 7PCBs, presuming that content of 7PCBs in the commercial Aroclor 1254 mixture is 31.2 %. According to Risso *et al.* (2016), it is possible to establish quantitative relationship between any single Aroclor mixture and the sum concentration of the PCB congeners (Risso, Magherini, Ottonelli, *et al.*, 2016).

Presented results exceed the limit values of 0.014 μg/L for the class III water established by ECE (Economic Commission for Europe) Standard Statistical Classification of Surface Freshwater Quality for the Maintenance of Aquatic Life (UN/ECE, 1992). Previously mentioned, Directive on classification of the surface water and water bodies categorization in Republika Srpska (Directive, 2001) set limit for the PCBs for class II water at 0.02-0.04 μg/L (expressed as sum of PCBs). Concentrations of PCBs at all five locations exceeded this limit. The total concentration of PCBs (ΣPCBs) determined in the Yangtze River, ranged in 0.04–11 ng/L, and was considered low to moderate polluted (Cui, Dong, Huang, *et al.*, 2020). Comparing to this, water in Lake Modrac can be considered as highly polluted. There is no other data on presence of the organic pollutants in the Lake Modrac, so we could not compare our results with similar studies. Additional surveys are urgently needed in order to evaluate ecological status of the accumulation Lake Modrac. Correlations between metals and PCBs concentrations were not statistically significant.

CONCLUSION

This is the first survey on levels of organic pollutants namely polychlorinated biphenyls (PCBs) in the accumulation Lake Modrac. The content of PCBs determined in this study exceeded the limit values established by regulatory bodies in EU and in BiH. Comparing values to test results obtained in other countries, water in the accumulation Lake Modrac can be considered as highly polluted. Regarding the content of heavy metals, the most abundant metals at all five sampling locations were Pb, Ni and Hg. Since the results from this study were higher than those reported from other authors for lakes in the region, it can be concluded that Lake Modrac evidently reflects some anthropogenic sources of pollution (possibly coal mining, industry, agricultural and domestic sources). Although, some previous surveys on presence of the heavy metals in Lake Modrac were conducted, results of this study are valuable addition to the existing data enabling the analysis of temporal trend in this area of BiH. Results of this study pointed out necessity of conducting more detailed monitoring of the ecological status of the accumulation Lake Modrac. This especially because of the recorded high content of the organic pollutants, that can pose significant health risk for humans.

REFERENCES

Abraxis, www. https://abraxis.eurofins-technologies.com/media/4640/pcbs-higher-chlorinated-elisa-user-guide-530041.pdf (13/6/2021)

Ashrap, P., Meeker, J. D., Sánchez, B. N., Basu, N., Tamayo-Ortiz, M., Solano-González, M., Mercado-Garcia, A., Téllez-Rojo, M. M., Peterson, K. E., Watkins, D. J. (2020). *In utero* and peripubertal metals exposure in relation to reproductive hormones and sexual maturation and progression among boys in Mexico City. *Environmental Health, 19*, 124, https://doi.org/10.1186/s12940-020-00672-0

BAS EN 13805:2015. (2015). Foodstuffs-determination of trace elements-pressure digestion. Sarajevo: Institute for Standardization of Bosnia and Herzegovina-ISBIH. (*In English)*

BAS EN 15586:2005. (2005). Water quality-Determination of trace elements using atomic absorption spectrometry with graphite furnace. Sarajevo: Institute for Standardization of Bosnia and Herzegovina-ISBIH. *(In English*)

Cui, X., Dong, J., Huang, Z., Liu, C., Qiao, X., Wang, X., Zhao, X., Zheng, B., Shen, J. (2020). Polychlorinated biphenyls in the drinking water source of the Yangtze River: characteristics and risk assessment. *Environmental Sciences Europe, 32*, 29, https://doi.org/10.1186/s12302-020-00309-6

Decision on characterization of surface and underground waters, reference conditions and parameters for the estimation of the water status and water monitoring in Federation of BiH. (2014). *Official Gazette of FBiH*, 1/14

Directive on classification of the surface water and water bodies categorization in Republika Srpska. (2001). Official Gazette of Republic of Srpska, 42/01

Directive on waterbodies categorization (1967). *Official Gazette of SRBiH*, 42/67

Đonlagić, N., Odobašić, A., Bratovčić, A. (2007). Influence of Agriculture on Water Quality: Significance of Heavy Metals Monitoring. *Agriculturae Conspectus Scientificus, 72 (4)*, 377-381.

Đozić, A., Alihodžić, H. (2019). *Analysis of the physicochemical parameters and presence of the heavy metals in the lake Modrac and River Spreča.* Center for Ecology and Energy, Tuzla (*In Bosnian)*

[EC] European Commission. (1975). Council Directive 75/440/EEC concerning the quality required of surface water intended for the abstraction of drinking water in the Member States. *Official Journal of the European Communities*, L194/26.

[EC] European Commission. (2000). Directive 2000/60/EC of the European Parliament and of the Council of 23 October 2000 establishing a framework for Community action in the field of water policy. *Official Journal of the European Union*, L327

[EC] European Commission. (2013). Directive 2013/39/EU of the European Parliament and of the Council of 12 august 2013 amending directives 2000/60/EC and 2008/105/EC as regards priority substances in the field of water policy. *Official Journal of the European Union*, L226/1.

Harman, C., Grung, M., Djedjibegovic, J., Marjanovic, A., Fjeld, E., Veiteberg Braaten, H. F., Sober, M., Larssen, T., Ranneklev, S. B. (2018). The organic pollutant status of rivers in Bosnia and Herzegovina as determined by a combination of active and passive sampling methods. *Environmental Monitoring and Assesment, 190(5), 283, https://doi.org/10.1007/s10661-018-6667-6*.

Harman, C., Grung, M., Djedjibegovic, J., Marjanovic, A., Sober, M., Sinanovic, K., Fjeld, E., Rognerud, S., Ranneklev, S. B., Larssen, T. (2013). Screening for Stockholm Convention persistent organic pollutants in the Bosna River (Bosnia and Herzogovina). *Environmental Monitoring and Assesment, 185(2)*, 1671-1683.

Institute for chemical engineering (2016). Investigation of the water quality of accumulation Lake Modrac in February 2016. (*Unpublished results, in Bosnian).*

Kelley, A. S., Banker, M., Goodrich, J. M., Dolinoy, D. C., Burant, C., Domino, S. E., Smith, Y. R., Song, P. X. K., Padmanabhan, V. (2019). Early pregnancy exposure to endocrine disrupting chemical mixtures are associated with infammatory changes in maternal and neonatal circulation. *Scientific Reports, 9*, 5422, https://doi.org/10.1038/s41598-019-41134-z

Klánová, J., Kohoutek, J., Kostrhounová, R., Holoubek, I. (2007). Are the residents of former Yugoslavia still exposed to elevated PCB levels due to the Balkan wars? Part 1: Air sampling in Croatia, Serbia, Bosnia and Herzegovina. *Environment International. 33(6)*, 719-726.

Kumar, M., Kumar Sarma, D., Shubham, S., Kumawat, M., Verma, V., Prakash, A., Tiwari, R. (2020). Environmental Endocrine-Disrupting Chemical Exposure: Role in Non-Communicable Diseases. *Frontiers in Public Health, 8, https://doi.org/10.3389/fpubh.2020.553850*

Lee, J., Lee, S., Yu, S., Rhew, D. (2016). Relationships between water quality parameters in rivers and lakes: BOD5, COD, NBOPs, and TOC. *Environmental Monitoring and Assesment, 188*, 252, DOI 10.1007/s10661-016-5251-1.

Li, Q.Q., Loganath, A., Chong, Y.S., Tan, J., Obbard, J.P. (2006). Persistent organic pollutants and adverse health effects in humans. *Journal of Toxicology and Environmental Health, Part A, 69 (21)*, 1987-2005.

Nwadiuto Amadi, C., Nkeiruka Igweze, Z., Ebere Orisakwe, O. (2017). Heavy metals in miscarriages and stillbirths in developing nations. *Middle East Fertility Society Journal, 22,* 91–100.

Omeragic, E., Radosevic, D., Causevic, A., Marjanovic, A., Djedjibegovic, J., Sober, M. (2021). Exposure assessment and risk characterization of metals intake through consumption of wine by population of winemakers in Bosnia and Herzegovina and Croatia. *Journal of Food and Nutrition Research, (published online first https://www.vup.sk/en/index.php?mainID=2&navID=34&version=2&volume=0&article=2222)*

Reports JP Spreča on results of the water analysis in the accumulation lake Modrac (2017). https://spreca.com/izvjestaji/ (12/6/2021) (*in Bosnian).*

Sibal, L.N., Espino, M.P. (2018). Heavy metals in lake water: a review on occurrence and analytical determination. *International Journal of Environmental Analytical Chemistry*, DOI: 10.1080/03067319.2018.1481212

Strategy of the protection of the accumulation Lake Modrac (2012). http://www.vladatk.kim.ba/Ministarstva/MPVS/2013/Strategija_zastite_akumulacije_Modrac.pdf (10/6/2021) (*in Bosnian)*

Street, M. E., Angelini, S., Bernasconi, S., Burgio, E., Cassio, A., *et al.* (2018). Current Knowledge on Endocrine Disrupting Chemicals (EDCs) from Animal Biology to Humans, from Pregnancy to Adulthood: Highlights from a National Italian Meeting *International Journal of Molecular Science, 19 (6),* 1647, https://doi.org/10.3390/ijms19061647.

UN/ECE. (1992). ECE Standards Statistical Classification of Surface Water Quality for the Maintenance of Aquatic Life" United Nation Economic and Social Council, Statistical Commission and Economic Commission for Europe. Conference of European Statisticians. CES/733, 13 April 1992.

Risso, F., Magherini, A., Ottonelli, M., Magi, E., Lottici, S., Maggiolo, S., Garbarino, M., Narizzano, R. (2016). A comprehensive approach to actual polychlorinated biphenyls environmental contamination. *Environmental Science and Pollution Research, 23(9),* 8770-8780.

Warmer, H. van Dokkum, R. (2002). *Water pollution control in the Netherlands-Policy and practice* 2001(2002.009). RIZA, Netherlands

Summary/Sažetak

Akumulaciono jezero Modrac je od posebnog značaja kao izvor pitke vode za stanovnike Tuzle i nekoliko okolnih naselja. Najznačajniji izvor kontaminacije akumulacije Modrac organskim polutantima predstavljaju otpadne vode iz domaćinstava i industrije. U ovoj regiji, većina naselja nema adekvatne sisteme odvoda otpadnih voda niti postoje postrojenja za njihov tretman. Ostali potencijalni izvori polutanata su industrijska postrojenja od kojih su najznačajniji rudnici (Banovići, Đurđevik), metalna i drvna industrija, fabrike za proizvodnju plastike, te skladišta nafte i naftnih derivata. Nekoliko prethodno provedenih istraživanja u regionu ukazalo je na značajno prisustvo postojanih organskih polutanata i teških metala, pa je cilj ove studije bio da se provede ispitivanje prisustva odabranih anroganskih (Cd, Cr, Cu, Hg, Ni, Pb i As) i organskih polutanata u vodi akumulacionog jezera Modrac, BiH. Sadržaj polihloriranih bifenila (PCB) određen ELISA testom kretao se u rasponu od 3,23-6,19 ppb (suma 7PCB), dok su najveće koncentracije metala (određene primjenom atomske apsorpcione spektrometrije - grafitne tehnike i živinim analizatorom) na svih pet lokacija uzorkovanja registrovane za olovo (6,79-36,58 µg/l), nikl (5,81-10,43 µg/l) i živu (1,08-6,10 µg/l).

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

2021

57

Original Scientific Article

Print ISSN: 0367-4444
Online ISSN: 2232-7266

DOI: https://doi.org/10.35666/2232-7266.2021.57.03

UDC: 613.84 [546.48:546.47]

17-26

Smoking effect on the cadmium and zinc concentration in smokers and nonsmokers

Pepić, E.[a,*], Šečić, D.[a], Lepara, O.[a], Kapić-Pleho, A.[a], Kurtagić-Pepić, E.[b], Mušanović, J.[a], Metović, A.[a], Džubur, A.[c], Naser, N.[d], Panjeta, M.[c], Alić-Džubur, A.[a], Voljevica, A.[a]

[a]Medical Faculty, University of Sarajevo, Čekaluša 90, Sarajevo, Bosnia and Herzegovina
[b]Institute for Public Health of Canton Sarajevo, Dr MustafePintola 1, Sarajevo, Bosnia and Herzegovina
[c]Clinical Center of Sarajevo University, Bolnička 25, Sarajevo, Bosnia and Herzegovina,
[d]Polyclinic "Dr. Nabil", Bulevar MešeSelimovića 2b, Sarajevo, Bosnia and Herzegovina

Article info
Received: 21/06/2021
Accepted: 15/12/2021

Keywords:
cadmium
zinc
smokers
former smokers
nonsmokers

***Corresponding author:**
Esad Pepić
E-mail: esad.pepic@mf.unsa.ba
Phone: +387 62 216 151

Abstract: Studies have shown that cigarette smoking affects the accumulation of some heavy metals in certain tissues and metabolism of essential elements. The aim of the study was to determine the differences in the concentrations of cadmium in the blood and urine and zinc in the urine of smokers and ex-smokers in relation to non-smokers, and to determine the possible influence of cadmium concentration on zinc excretion as an essential element. The study included 106 subjects. Subjects were regular smokers (n=51), ex-smokers (n=38) and non-smokers (n=17). Atomic absorption spectrophotometry (AAS) with an electrothermal atomizer was used to determine cadmium. Zinc was determined by AAS with a flame atomizer. There was a significant difference in the values of cadmium in the blood between the groups: smokers and non-smokers ($p<0.001$), smokers and ex-smokers ($p <0.001$), and between ex-smokers and non-smokers ($p = 0.045$). There is a significant positive and strong correlation in the level of zinc and cadmium in urine per gram of creatinine, and as the level of cadmium increases, the level of zinc also increases (rho=0.781; p=0.001). The data indicate that cigarette smoking has been shown to be a factor that can increase cadmium levels to an extent that will significantly increase zinc excretion, or its increased loss.

INTRODUCTION

Cigarette smoking, which produces chemical compounds that have a very detrimental effect on the health of smokers and people in their vicinity, is a widespread bad habit among people of all ages. Harmful effects on the human body are based on the fact that cigarette smoke, in addition to nicotine, contains more than 5300 other ingredients, of which more than 70 are carcinogenic (Talhout, Schulz, Florek, *et al.* 2011). Heavy metals, such as cadmium, mercury, chromium, lead, antimony, nickel, and zinc, are important categories of carcinogenic ingredients in tobacco smoke. After entering the body, toxic metals have a long biological lifespan, which affects the accumulation of some heavy metals in certain tissues and the metabolism of essential elements, with changes in their values in biological material, primarily blood, urine, hair, and nails (Alrobaian, Arida, 2019).

The role of these metals in the pathophysiology of diseases caused by cigarette smoking is still poorly understood. New data suggest that changing the homeostasis of these metals in the human body by cigarettes smoking plays a crucial role in the development of several diseases, primarily respiratory and cardiovascular, and malignant cell alteration (Bernhard, Rossmann, Wick, 2005). Cadmium as an element has no physiological or biological role in the human body. The most common pathways when cadmium enters the body are inhalation and intake of food or water that contains large amounts of cadmium. Cadmium poisoning rarely causes death and leads to cancer and toxicity of organs such as skeletal, urinary, reproductive, cardiovascular, nervous, and respiratory systems. Most commonly it causes nausea and vomiting. The rest of the cadmium accumulates well in the body, and if metallothionein does not remove it from the body

in a short period, it can remain in humans for up to several decades. The decay time of cadmium in the kidneys is 6 to 38 years, while in the liver, it is 9 to 18 years, and the reason is that cadmium has no function in the body, and the body does not consume it (Prashanth, Kattapagari, Chitturi, *et al.* 2015).

The effect of cadmium on the body is to increase the activity of proinflammatory cytokines and causes diseases in many tissues, including the lungs and kidneys (Alkan, Cakmak, Karis, 2014). In addition, cadmium negatively affects the process of DNA replication and reparation and increases the possibility of error, *i.e.*, it acts as a mutagen. With chronic exposure to cadmium, this mutagenic effect multiplies and becomes carcinogenic, which often cannot be corrected by a subsequent increase in the presence of zinc (Lützen, Liberti, Rasmussen, 2004). Previous studies have shown that smokers have higher cadmium concentrations in urine, blood, hair, and tissues than non-smokers. While urinary cadmium concentrations correlate with chronic exposure to tobacco smoke, blood cadmium concentrations provide information on recent cadmium exposure (Richter, Faroon, Pappas, 2017).

Zinc is an essential element necessary for the proper growth, development, and functioning of the human body. This mineral is found in almost every phase of activation of antioxidant enzymes and is necessary for participation in important biological functions. It has a high antioxidant potential and is an integral part of 300 enzymes, as cofactor it participates in the synthesis and breakdown of carbohydrates, lipids, proteins, and nucleic acids. The human body contains 2-3 grams of zinc, and it is after iron the second trace element in the human body (Prashanth, Kattapagari, Chitturi, 2015). Zinc is essential for the creation, development, and maintenance of the immune system involved in the healing wounds, injuries, and burns. It needs the conversion of vitamin A in its active form to play an important role in the production of the hormone testosterone and the balancing insulin in the human body. With its antioxidant properties, zinc participates in the body's defense against free and harmful radicals. It is also needed for a normal sense of taste and smell and normal growth and development during pregnancy, childhood, and adolescence (Osredkar, Sustar, 2011). Less than 0.5% of the total body amount of zinc is found in plasma, while most is stored mainly in muscles and bones (approximately 90%) and the liver, teeth, hair, skin, leukocytes, testes, and others. Most of the plasma zinc binds up to proteins, about 50% to albumins (Al-Assaf, 2010). It is known that even the slightest change in zinc concentration can lead to the development and progression of many mild and severe acute and chronic diseases. Zinc deficiency can negatively affect the processes of genetic mutation and carcinogenesis and increase oxidative stress (Proudfoot, McPherson, Kolb, 2011; Dhawan, Chadha, 2010).

Data on the concentration of zinc in tobacco smoke are sparse and inconsistent. The content of cadmium in the topsoil (0-10 cm) varied from 0.01 to 16.9 μg/g. The concentrations of cadmium in cigarettes range from 0.5 to 3.5 μg/g, with a mean level of 1.7 μg/g. These are very high levels compared to those in food that are normally below 0.05 μg/g. Zinc concentration, analysed in 12 American cigarette brands, range from 16.8 to 30.5 μg/g (Chiba, Masironi, 1992). During combustion, less zinc is generated from tobacco into smoke than cadmium, indicating a negligible zinc concentration compared to cadmium in common cigarette smoke particles (Fresquez, Pappas, Watson, 2013).

The high positive correlation or interdependence between the values of cadmium and zinc in urine can be explained by the metabolic antagonism of cadmium and zinc, which arises on the principle of competition for the same carrier. In this case, it is metallothionein, a low molecular weight cytoplasmic protein for which cadmium and zinc compete for cystine sites (Goyer, 1997). This competition goes to the account of zinc if the value of cadmium is high, which explains this strong, cross-correlation and which causes an increase in the loss of zinc. Cadmium bound to metallothionein in renal and liver, epithelial cells, is not toxic, but cadmium bound to plasma metallothionein is toxic to the renal tubules as it is excreted in the urine, as confirmed by Chan and Cherian (1993) in an animal model.

The aim of the study was to determine the difference in the values of cadmium and zinc in the biological material of smokers in relation to ex-smokers and non-smokers and to examine the interdependence of cadmium and zinc levels in smokers, ex-smokers, and non-smokers.

EXPERIMENTAL

Subjects

The research included 108 participants living in Sarajevo Canton., According to the inclusion criteria, the participants were classified into three groups. The first group consists of regular smokers (n = 51), the second group consists of ex-smokers (n = 38), and the third group of participants who have never consumed tobacco (n = 17). During the study, two subjects were excluded due to extremely high levels of cadmium from the first and second group. The smoking experience of ex-smokers was 30.94 ± 18.46 pack-year, while for current smokers, this experience was 32.78 ± 17.70 pack-year.

All participants had a detailed medical interview. Individual questionnaires included data on gender, age, smoking status, height and weight, and body mass index (BMI), eating habits (consumption of fish, meat, alcohol, and coffee), existence and a number of amalgam dental fillings, type of occupation (office or more physical work), education, the presence of an acute illness or a chronic illness. For smokers, data on the length of smoking experience with average daily smoked cigarettes were taken, and for the group of former smokers, data on the length of the smoking experience, average daily smoked cigarettes, and the length of non-tobacco consumption.

Criteria for inclusion in the study were: participants who are not occupationally exposed to heavy metals, participants who do not take trace element supplements as a dietary supplement, voluntary consent to participate in research, participants who do not take other intoxicants, permanent residence in Canton Sarajevo for at least 20 years, and consumption at least10 cigarettes

daily for smoking participants. Exclusion criteria were: consumption of less than 10 cigarettes per day in a group of smokers, consumption of less than 10 cigarettes by ex-smokers during smoking, occupational exposure to heavy metals, data on drug use, taking zinc supplements as dietary ones, existence of metal implants, residence in Sarajevo Canton for less than 20 years. The study was conducted with the approval of the Ethics Committee of the Faculty of Medicine, University of Sarajevo (1324-AS/11) in accordance with the recommendations contained in the Declaration of Helsinki on Biomedical Research Involving Human Subjects as revised in 2013.

Sample analysis

After a medical interview and physical examination, the subjects had their blood taken for laboratory tests by puncturing the cubital vein. Serum was extracted from blood samples after coagulation and centrifugation for 10 minutes at 4000 rpm and stored until the required results were obtained. A blood sample (10 cm^3) was taken between 8.00 and 9.30 am using a vacutainer system. The vacutainer system consists of a plastic cylinder, a disposable needle, and a vacuum tube. Contact with the needle, vacutainer walls, and stopper was avoided during blood collection. The blood vacutainer is clogged with care to avoid contamination. The blood sample was analyzed immediately or left for up to 2 days at + 4ºC. If the analysis was performed over several days, then the samples were stored at -20ºC. Before analysis, the blood was stirred for half an hour on a ROLLER. EU-certified blood (serum) BCR194L, BCR194N, BCR 194H was used as a control. For the first sample, 20 cm^3 of urine was taken from the patient once in a plastic container made of chemically inert material. The sample was labeled and left at -20°C.

Determination of heavy metal concentration in a sample

Determination of heavy metal content was performed in the Laboratory for the Toxicology of the Public Institution Institute for Occupational Medicine of Sarajevo Canton and the Institute for Public Health of Sarajevo Canton by atomic absorption spectrophotometry (AAS). This is the most commonly used analytical technique for determining metals in samples. The metals being analyzed absorb radiation of a certain wavelength, whereby excitation occurs at the level of electrons and their transition to higher energy levels. Radiation from an external source of energy corresponds to the difference between the ground and excited states of atoms or ions. For the same electron transition, the energy of the emitted photon is equivalent to the energy of the absorbed, that is, the wavelength of the emitted is equal to the wavelength of the absorbed radiation. The advantage of AAS is the speed of analysis, simplicity, relatively low cost of instruments, and favorable sensitivity and selectivity for many elements. Limitations are the difficulty in determining multiple elements at the same time because each element requires a corresponding hollow cathode lamp (Jignesh, Vineeta, Abhay, 2012).

Determination of cadmium levels in blood and urine

Cadmium levels were determined by Graphite Furnace Atomic Absorption Spectroscopy (GFAAS). The main instruments and devices used in the measurement were:

- Atomic absorption spectrophotometer Perkin Elmer Model, USA, AAnalyst 600. THGA technique (thermally heated graphite cuvette),
- Autosampler model AS-800;
- WinLab 32 software.

Other instruments used:

- Nahita centrifuge, model 2690
- Centrifuge EBA 20 (HettichzentrifugenTuttlingen, Germany)
- Mixer Vortex - 2 - Genie, Scientific Industries BOHEMIA N.Y. 11716 USA Laboratory Equipment Model No. G-560E
- Demineralized water machine Sartorius Arium 63316, Sartorius Arium 611UV, Sartorius Tank 613 APV31 30 L, 10 bar, 90°C.

Preparation of blood and urine for the cadmium level measurement

Due to its low affinity for the measured heavy metals, K2-EDTA (Ethylenediaminetetraacetic Acid) was used as a blood anticoagulant. Before determining the cadmium levels in the sample, it was necessary to create a calibration curve. A pool of blood and a pool of urine were added to all solutions for the calibration curve when determining cadmium in whole blood and urine. All solutions were prepared in deionized water of very high purity with 0.2% HNO_3. Calibration is performed by a four-point instrument from two standard solutions 1 µg/L and 5 µg/L. Calibration points (standards) were: 0.2 µg/L, 0.5 µg/L, 1 µg/L and 5 g/L. Each point was read three times, which was the basis for the mean value calculation. The minimum number of points on the calibration curve was four, and the correlation coefficient must be $\geq$ 0.995. Standards have been developed whose values cover the possible range of cadmium levels in the samples. The sample amount is 10 µl. HNO_3 precipitated blood proteins. An aliquot of the sample was injected into an AAS graphite cuvette, and the cadmium absorbance was measured at 228.8 nm. Cadmium pyrolysis (thermal decomposition) takes place at 700°C and atomization at 1550°C. The recommended atomization time was 3 seconds (max. 10 sec.). The cadmium level is calculated using a calibration curve made by the standard addition of cadmium to the blood. They were read in three measurements from which the mean value was taken.

Method for determining zinc in urine

The level of zinc in the urine was determined by atomic absorption spectrophotometry with a flame atomizer. The measurement was performed on a Varian device, model: SpectrAA 110 Atomic Absorption Spectrometer. The zinc standard used in the analysis was 1 mg/ml, manufactured by Panreac. The principle of this method is to determine the concentrations of zinc in different samples by measuring the absorbance of the sample.

When the sample burns, an atomization process occurs, and the resulting zinc metal atoms are exposed to line radiation of a specific wavelength (213.9 nm). The photodetector detects the change in radiation intensity, and the instrument software translates the radiation intensity into the metal concentration in the sample. Prior to each use, the instrument should be calibrated with a pre-prepared concentration series of standard zinc solutions. After calibration of the instrument, it is possible to determine the zinc in the samples. The samples are homogenized and aspirated using a plastic tube in such a way that the tube is immersed in the sample solution where the device automatically sucks up the solution continuously. Since the device allows multiple consecutive measurements and analysis of the same homogeneous sample quickly, based on a pre-set sample, three measurements are performed, and the average value is calculated.

Adjustment of measurements and standards in determining the value of zinc:

- Wavelength: 213.9 nm.
- Separation wavelength: 1.0 nm
- Lamp current: 5.0 mA
- Number of replica samples and standards: 3
- Time measurement: 5s
- Flame type: air / acetylene
- Air flow rate: 3.5 L / min.
- Acetylene flow rate: 1.5 L / min.

The sample is prepared for measurement by taking a 400 μL urine sample in a glass tube and adding 1600 μL of purified water and stirring. The blank is made of ultrapure water that does not contain a sample. The flame atomizer needs to be purified with 2% HNO_3, and between individual measurements, ultrapure water is used for rinsing. After preparation and homogenization, the samples are measured. The device allows multiple consecutive measurements of the same sample quickly, which contributes to greater measurement accuracy. Zinc concentrations in urine samples were calculated using a calibration curve and expressed in mg/L.

Other analyzes

Each participant was determined for a complete blood count and the concentration of creatinine in the urine. The concentration of creatinine in urine is the best indicator of urine concentration, and its determination was necessary to express the concentration of heavy metals so that the concentration of heavy metal is expressed in micrograms per gram of creatinine in urine (μg/g creatinine in urine - μg/g Ucr). Thus, the concentrations of heavy metals can be compared with each other regardless of the concentration of urine, which is an excellent advantage over the volume expression of heavy metals in μg/L of urine. This is a convenient method when the metal concentration is not determined in 24-hour urine.

Statistical analysis

For statistical data processing *Microsoft Excel 2013* and *IBM SPSS Statistics 20* were used. The results are presented as the median, with the first and third quartile values (Q1 and Q3) or as an arithmetic mean with the corresponding standard deviation, depending on the data distribution. The values were compared using Student t-test or non-parametric test (Man Whitney test). The Kruskall-Wallis test or analysis of variance (ANOVA) was used to compare the values of more than two groups, depending on the data distribution. The Spearman's coefficient correlation analysis was used to correlate data. A significance level of 5% was used to determine statistical differences.

RESULTS AND DISCUSSION

The basic characteristics of respondents, smokers, non-smokers and ex-smokers are shown in Table 1. These data represent characteristics of age, body mass index (BMI), gender, type of job (intellectual, or physical), and the presence of a chronic disease.

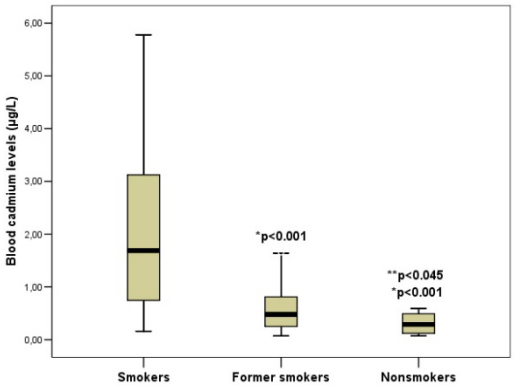


Figure 1: Concentration of cadmium in the blood *smokers in comparison to non-smokers and ex-smokers **ex-smokers in comparison to non-smokers

The analysis of cadmium in the respondent's blood (Figure 1) showed the highest level in smokers with a median of 1.74 μg/L (0.74 μg/L; 3.19 μg/L), followed by ex-smokers 0.44 μg/L, (0.22 μg/L; 0.64 μg/L), while in non-smokers this value was 0.29 μg/L (0.10 μg/L; 0.50 μg/L).

There was a difference in the values of cadmium in the blood between the groups: smokers and non-smokers ($p<0.001$), smokers and ex-smokers ($p <0.001$), and between ex-smokers and non-smokers ($p = 0.045$).

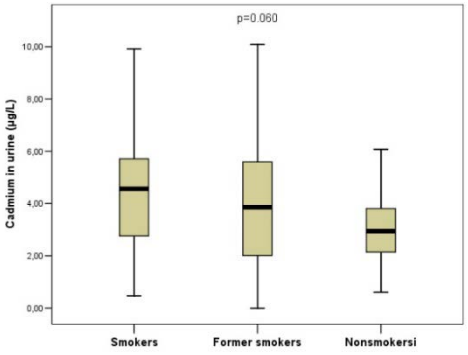


Figure 2: Cadmium volume values in urine

The volume values of cadmium in the urine of the respondents (Figure 2) were found to be at the highest levels in smokers with a median of 4.56 μg/L (2.68 μg/L; 5.80 μg/L), followed by ex-smokers 4.22 μg/L, (2.01 μg/L; 5.79 μg/L), while in non-smokers this value was 2.94 μg/L (1.77 μg/L; 3.90 μg/L).

Table 1: Patients characteristics

		Smokers	**Former smokers**	**Non-smokers**
	Age (years)	47.80 ± 5.68	50,57 ± 6.16	48,88 ± 5.60
	BMI (kg/m^2)	27.2 ± 4.6	28.6 ± 4.3	27.2 ± 4.6
Gender	Male (%)	47.1	55.3	41.2
	Female (%)	52.9	44.7	58.8
Occupation	Intellectual work (%)	51.0	63.0	59.0
	Physical work (%)	49.0	37.0	41.0
Health status	No concomitant disease (%)	49.0	47.4	94.1
	Chronic diseases (%)	45.1	44.7	5.9
Smoking experience (pack-year)		32.78 ± 17.70	30.94 ± 18.46	-

*For age, BMI and smoking experience period data expressed in mean±SD

The differences between the tested groups are indicative, but not significant (p=0.060).

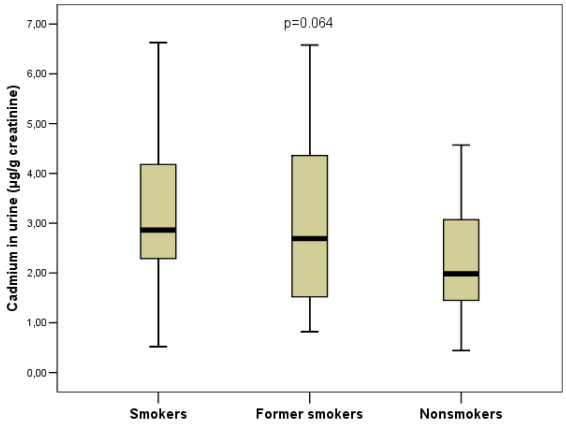


Figure 3: Cadmium levels in urine per gram of creatinine

Cadmium values per gram of creatinine in urine (Figure 3) indicate that the highest level was found in smokers with a median of 2.86 µg/L (2.22 µg/L; 4.24 µg/L), followed by ex-smokers 2.56 µg/L, (1.42 µg/L; 4.36 µg/L), while in non-smokers this value was 1.98 µg/L (1.43 µg/L; 3.24 µg/L).

Differences between the examined groups did exist, but were not significant (p=0.064).

Volume zinc concentrations and zinc concentrations per gram of creatinine in urine did not show significant differences between the tested groups (p=0.909; p=0.877) (Table 2).

In the group of smokers, a significant positive correlation was found between the concentration of cadmium in urine per gram of creatinine (µg/g creatinine) and the duration of smoking (rho=0.323; p<0.05). In the same group, a significant positive correlation was found between the concentration of cadmium in the blood with the number of pack-year (rho=0.293; p<0.05) as well as the concentration of cadmium in urine per gram of creatinine with the number of pack-year (rho=0.300; p<0, 05) (Table 3) A strong positive correlation was also found between the concentration of cadmium in the blood and the number of cigarettes smoked per day (rho=0.485; p<0.01) (Table 3).

Table 2: Median values and 25 and 75 percentiles for level of cadmium in blood and urine and zinc in urine

Metal/sample type	Groups	Percentiles		
		25th	Median	75th
Cadmium/Blood (µg/L)	Smokers	0.75	1.74	3.20
	Ex-smokers	0.22	0.44	0.64
	Non-smokers	0.11	0.29	0.50
Cadmium/Urine (µg/L)	Smokers	2.68	4.56	5.81
	Ex-smokers	2.01	4.22	5.79
	Non-smokers	1.77	2.94	3.91
Cadmium/Urine Creatinine (µg/g)	Smokers	2.23	2.86	4.24
	Ex-smokers	1.42	2.56	4.36
	Non-smokers	1.44	1.98	3.25
Zn/Urine (µg/L)	Smokers	189.8	375.5	579.3
	Ex-smokers	238.0	353.5	525.5
	Non-smokers	254.0	333.0	532.5
Zn/Urine Creatinine (µg/g)	Smokers	196.6	249.5	370.3
	Ex-smokers	171.6	254.2	389.3
	Non-smokers	222.8	295.9	347.1

Correlations between cadmium levels in blood with smoking duration, cadmium levels in urine with smoking duration, number of pack-year, number of cigarettes per

day, cadmium per gram of creatinine values in urine with number of cigarettes per day, zinc levels in urine with smoking duration, number of pack-year, number of cigarettes per day, zinc per gram of creatinine values in urine with smoking duration, number of pack-year, number of cigarettes per day did not show statistical significance (Table 3).

Table 3: Correlations between the concentration of cadmium in blood and urine, zinc in urine with years of smoking duration, number of pack-year and daily smoked cigarettes in a group of smokers

Variable	Smoking duration	Number of pack- year	No. of cigarettes per day
Cadmium Blood	Rho= 0.022	**Rho= 0.293***	**Rho= 0.485****
Cadmium Urine	Rho= 0.072	Rho= 0.144	Rho= 0.093
Cadmium Urine/Urine Creatinine	**Rho= 0.323***	**Rho= 0.300***	Rho= 0.159
Zinc Urine	Rho= -0.172	Rho= -0.074	Rho= 0.014
Zinc Urine/Urine Creatinine	Rho= -0.051	Rho= 0.015	Rho= 0.090

Level of significance:
*p<0.05
**p<0.01

There is a statistically significant positive and strong correlation in the level of zinc and cadmium in urine per gram of creatinine, and as the level of cadmium increases, the level of zinc also increases (rho=0.781; p=0.001)

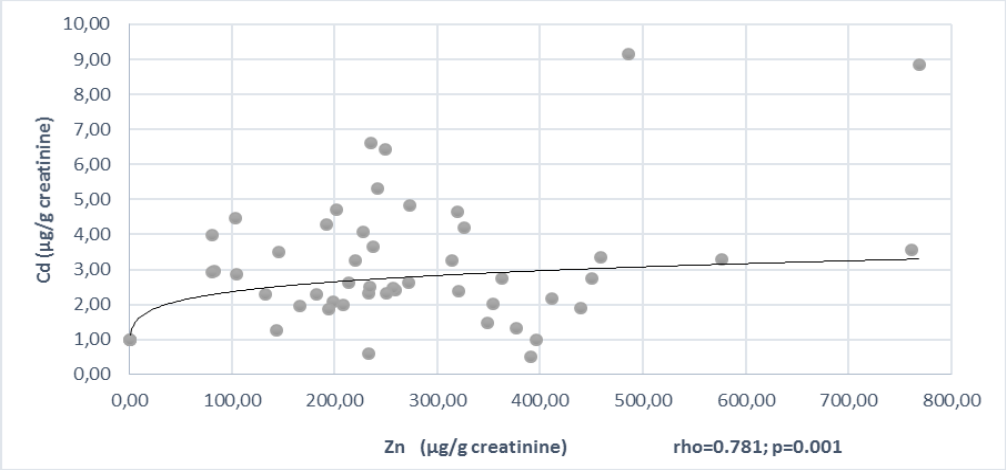


Figure 4: Correlation of zinc and cadmium levels in the urine of smokers per gram of creatinine

Previous studies have shown that cadmium absorption is most effective by the respiratory system (Nordberg, Kjellstrom, Nordberg, 1985). So perennial smoking represents a chronic exposure to low values of cadmium. Olsson, Bensryd, Lundh*et al.* (2002) conducted a study on 105 participants, of whom 26 participants were ex-smokers and 79 who had never consumed tobacco. They determined the level of cadmium in blood and urine of all subjects. They found higher blood cadmium values in ex-smokers, compared to non-smokers. Our study also found a statistically significant difference in cadmium values between ex-smokers and non-smokers (p=0.045). Since the half-life of cadmium in the blood is 2-3 months, we expected that the values will not be higher in ex-smokers compared to non-smokers, because according to some studies, the level of cadmium in the blood is a measure of short-term exposure (Birgisdottir, Knutsen, Haugen, *et al*., 2013). It is likely that the level of cadmium in the blood is a reflection not only of the short-term load of the body with cadmium, but also a consequence of long-term accumulation in the kidneys and liver (50-75% of the total cadmium content in the body), from where cadmium partially re-enters the blood, as shown in study conducted by Hoffmann, Krause, and Seifert (2001). Therefore, this increase in the levels of cadmium in blood of ex-smokers compared to non-smokers, and after several years of smoking cessation, could be taken into account in the biomonitoring cadmium exposure.

Our study found a statistically significant difference cadmium levels in blood between smokers and non-smokers (p<0.001). These results are consistent with the results of Anetor, Ajose, Anetor, *et al.* (2008) who conducted a study on 55 healthy smokers and 41 healthy non-smokers. They determined the level of cadmium and zinc in the serum, and assessed the health risks of the examined groups, primarily the risk of prostate cancer. The level of cadmium, in the aforementioned study by Anetor, *et al.* (2008), in the group of smokers was significantly higher compared to non-smokers. The same authors found a positive correlation between smoking history and blood cadmium concentration (p<0.05). Such large differences in the values of cadmium in the blood of smokers and non-smokers support the importance of respiratory absorption, as the most efficient form of intake of this heavy metal, which was confirmed by the studies by Elinder, Kjellstrom, Lind, *et al.* (1983).

Our study, in accordance with the described study, found a statistically significant difference between the measured values of cadmium in the blood between

smokers and ex-smokers ($p<0.001$). In addition, we found a positive correlation between the concentration of cadmium in the blood and smoking duration of in the group of smokers, so that long-term smokers had a higher concentration of cadmium in the blood. Smoking duration is expressed by the number of packs-year of cigarettes. We also found a positive correlation between the concentration of cadmium in the blood of smokers and the number of cigarettes smoked per day, which is consistent with previous study conducted by Anetor, Ajose, Anetor*et al.* (2008).

The results of the study we conducted are in accordance with the results of the study conducted by Massadeh, Gharibeh, Omari, *et al.* (2010) on a sample of 79 participants. These authors also found a positive correlation between blood cadmium levels and smoking duration as well as the number of cigarettes consumed per day. However, these authors, like the previous Anetor, *et al.*, did not include ex-smokers in their study. The results of the presented study on the level of cadmium in the blood are also consistent with the results of research by Birgisdottir, Knutsen, Haugen, et al. (2013) and Afridi, Kazi, Kazi, *et al.* (2011). Blood cadmium values were higher in subjects from all three groups (smokers, ex-smokers, non-smokers) with the chronic disease (chronic obstructive pulmonary disease-COPD, cardiovascular disease-CVD and malignancies; $p<0.05$). These results are confirmed by the studies of Anetor*et al.* (2008), and Afridi*et al.* (2010).

In our study, in addition to the volume value of cadmium levels in urine, we also determined its level expressed per gram of creatinine in urine (μg/g Ucr). The concentration of creatinine in urine is the best indicator of urine concentration and its determination is necessary in order to express the levels of heavy metals in this medium. This enables a more objective comparison of the values of heavy metals regardless of the concentration of urine, which is an advantage over the volume expression of the values of heavy metals, or in μg/L of urine. The values of cadmium in urine, expressed by this method, in our study were the highest in smokers, but without statistical significance ($p=0.06$). We also found a positive correlation between urine cadmium levels per gram of creatinine and smoking duration (number of pack-year), as well as the number of cigarettes consumed per day ($p<0.05$), which is in accordance with the studies of Bamgbose, Opeolu, Bamgbose (2007), and Olsson *et al.* (2002).

The results of our study are consistent with the results of a study by Benemann, Bromen, Lehmann, *et al.* (2004) (n=4551) conducted as part of heavy metal biomonitoring in the general population. This study found that a group of smokers with a smoking volume of 25 pack-year had an increase in urine cadmium levels of 45.5% expressed in μg/g Ucr. One of the possible causes of the increase in the amount of cadmium in urine among ex-smokers compared to non-smokers, and if it does not reach statistical significance in our study ($p=0.06$) is the biokinetics of cadmium, because this heavy metal is excreted by the kidneys upon release from its depots in the body. If we take into account that the half-life of elimination of cadmium from the body is long (20-30 years) and that the main route of elimination is through the kidneys and urine, it probably takes a longer period of time than the cessation of exposure, for the difference in concentrations between the group of ex-smokers and non-smokers to be insignificant, which we found in the study of Olson *et al.* (2002). A study conducted by Mutti, Corradi, Goldoni, *et al.* (2006) showed that cadmium and lead were still in measurable values in exhaled air condensation, years after smoking cessation.

In our study, the values of zinc in urine were determined. The volume values of zinc in urine and values of zinc in urine per gram of creatinine did not differ statistically significantly between the examined groups ($p=0.909$; $p=0.877$). However, there is a significant positive correlation between urine cadmium and zinc values expressed per gram of creatinine. As the level of cadmium increases, the level of zinc also increases ($rho= 0.781$; $p=0.001$). Such a high degree of correlation indicates that the increase in the content of cadmium in the body, or in its depots, which in turn increases the intensity of zinc loss by increased excretion, suggests increased loss of this essential element rather than its increased bioavailability as shown by studies by Afridi, Kazi, Kazi*et al.* (2010), and Lin, Caffrey, Chang *et al.* (2010).

Considering that zinc acts in the process of DNA replication, its stabilization, and in the processes of DNA reparation, as an integral and essential part of enzyme systems involved in these processes (Proudfoot *et al*, 2011), the effect of cadmium on its metabolism is even more significant.

In addition, zinc reduces oxidative stress and exhibits antioxidant activity. Zinc deficiency can adversely affect the processes of genetic mutation and carcinogenesis, and increase oxidative stress, as shown in studies conducted by Proudfoot, McPherson, Kolb*et al.* (2011), Dhawan, Chadha (2010) and Bertini, Decaria, Rosato, (2010). On the other hand, cadmium, as a zinc antagonist, negatively affects the process of DNA replication and reparation and acts as a mutagen. In chronic exposure to cadmium, this mutagenic effect multiplies and becomes carcinogenic, which often cannot be partially canceled by a subsequent increase in the presence of zinc [Clark and Kunkel (*2004*), Jin, Clark, Slebos*et al. (2003*), Waisberg, Joseph, Hale*et al. (2003*), Lützen, Liberti, Rasmussen (2004)].

CONCLUSION

The presence of a large number of harmful chemical elements in tobacco smoke such as cadmium has consequences on biological indicators, the so-called biomarkers of exposure. Biomarkers of cadmium exposure in blood and urine show significant shifts, especially in the blood of active smokers. This study also found a significant difference in cadmium values between ex-smokers and non-smokers. Therefore, this

increase in the level of cadmium in the blood of ex-smokers compared to non-smokers, and after several years of smoking cessation, could be taken into account in the biomonitoring of cadmium exposure. There is a significant positive correlation between cadmium and zinc values in urine expressed per gram of creatinine. Such a high degree of correlation indicates that the increase in the content of cadmium in the body, or in its depots, which in turn increases the intensity of zinc loss by increased excretion, suggests increased loss of this essential element rather than its increased bioavailability.

REFERENCES

Afridi, H. I., Kazi, T. G., Kazi, N., Kandhro, G. A., Baig, J. A., Jamali, M. K., Arain, M. B., Shah,A. Q. (2011). Interactions between cadmium and zinc in the biological samples of Pakistani smokers and nonsmokers cardiovascular disease patients. *Biological Trace Element Research*, 139(3), 257-268.

Afridi, H. I., Kazi, T. G., Kazi, N. G., Jamali, M. K., Arain, M. B., Sirajuddin, Baig, J. A., Kandhro, G. A.,Wadhwa, S. K., Shah, A. Q. (2010). Evaluation of cadmium, lead, nickel and zinc status in biological samples of smokers and nonsmokers hypertensive patients. *Journal of Human Hypertension*, 24(1), 34-43.

Al-Assaf, N. A. (2010). Determination of Serum Trace Elements Magnesium, Copper, Zinc, and Selenium in Asthmatic Patients by Atomic Absorption Spectrophotometry. *Journal of Al-Nahrain University*, 13(1), 20-25.

Alkan, F. A., Cakmak, G., Karis, D., Sağlam, Z. A., Saler, T., Temiz, L. U., Yenigün, M., Ercan, M. (2014). The evaluation of plasma viscosity and endothelial dysfunction in smoking individuals. *Clinical Hemorheology and Microcirculation*, 58(3), 403-413.

Alrobaian, M., Arida, H. (2019). Assessment of Heavy and Toxic Metals in the Blood and Hair of Saudi Arabia Smokers Using Modern Analytical Techniques. *International Journal of Analytical Chemistry*, 7125210.

Anetor, J. I., Ajose, F., Anetor, G. O., Iyanda,A. A., Babalola,O. O., Adeniyi,F. A. A. (2008). High cadmium / zinc ratio in cigarette smokers: potential implications as a biomarker of risk of prostate cancer. *Nigerian Journal of Physiological Sciences*, 23(1-2), 41-49.

Bamgbose, O., Opeolu, B. O., Bamgbose, J. T. (2007). Levels of Cadmium, Lead and Zinc in Urine of Randomly Selected Smokers and Non-Smokers Residents of Abeokuta City, Nigeria. *Research Journal* of *Applied*, 2(2), 192-197.

Benemann, J., Bromen, K., Lehmann, N., Marr, A. K. H. Jöckel, K. H. Umwelt-Survey 1998 Band VII: Arsen, Schwer - und Edelmetalle in Blut und Urin der Bevölkerung in Deutschland –Belastungsquellen und–pfade. *Institut für Medizinische Informatik, Biometrie und Epidemiologie Universitäts klinikum Essen, Umweltbundesamtes. Band VII: 38-101, Berlin 2004.*

Bernhard, D., Rossmann, A., Wick, G. (2005). Metals in cigarette smoke. *IUBMB Life*, 57(12), 805–809.

Bertini, I., Decaria, L., Rosato, A. (2010). The annotation of full zinc proteomes. *Journal of Biological Inorganic Chemistry*, 15(7), 1071-1078.

Birgisdottir, B. E., Knutsen, H. K., Haugen, M., Gjelstad, I. M., Jenssen, M. T. S., Ellingsen, D. G. Thomassen, Y., Alexander,J., Meltzer, H. M., Brantsæter, A. L. (2013). Essential and toxic element concentrations in blood and urine and their associations with diet: results from a Norwegian population study including high-consumers of seafood and game. *Science of the Total Environment*, 463-464, 836-44.

Chan, H. M., Cherian, M. G. (1993). Mobilization of hepatic cadmium in pregnant rats. *Toxicology* and *Applied Pharmacology*, 120(2), 308-314.

Chiba, M., Masironi, R. (1992). Toxic and trace elements in tobacco and tobacco smoke. *Bulletin of the World Health Organization*, 70 (2), 269-275.

Clark, A. B., Kunkel, T. A. (2004). Cadmium inhibits the functions of eukaryotic MutS complexes. *Journal of Biological Chemistry*, 279(52), 53903-53906.

Dhawan, D. K., Chadha, V. D. (2010). Zinc: a promising agent in dietary chemoprevention of cancer. *The Indian Journal of Medical Research*, 132(6), 676–682.

Elinder, C. G., Kjellstrom, T., Lind, B., Linnman, L., Piscator, M., Sundstedt,K. (1983). Cadmium exposure from smoking cigarettes - variations with time and country where purchased. *Environmental Research*, 32 (1), 220-227.

Fresquez, M. R., Pappas, R. S., Watson, C. H. (2013). Establishment of toxic metal reference range in tobacco from US cigarettes. *Journal of Analytical Toxicology*, 37(5), 298-304.

Goyer, R. A. (1997). Toxic and essential metal interactions. The *Annual Review* of *Nutrition*, 17:37-50.

Hoffmann, K., Krause, C., Seifert, B. (2001). The German Environmental Survey 1990/92 (GerES II): primary predictor of blood cadmium levels in adults. *Archives of Environmental Health, 56*, 374-379.

Jignesh S., Vineeta K., Abhay S., Vilasrao K. (2012). Analytical methods for estimation of metals. *International Journal* of *Research* in *Pharmacy* and *Chemistry*, 2(1), 146-149.

Jin Y. H., Clark, A. B., Slebos, R. J., Al-Refai, H., Taylor, J. A., Kunkel, T. A., Resnick, M. A., Gordenin D. A. (2003). Cadmium is a mutagen that acts by inhibiting mismatch repair. *Nature Genetics*, 34(3), 326-329.

Lin, Y.S., Caffrey, J. L., Chang, M. H., Dowling, N., Lin, J. W. (2010). Cigarette smoking, cadmium exposure, and zinc intake on obstructive lung disorder. *Respiratory Research*, 11(1), 53.

Lützen, A., Liberti, S. E., Rasmussen, L. J. (2004). Cadmium inhibits human DNA mismatch repair in vivo. *Biochemical and Biophysical Research Communications*, 321(1), 21-25.

Massadeh, A., Gharibeh, A., Omari, K., Al-Momani, I., Alomary, A.,Tumah, H., Hayajneh, W. (2010). Simultaneous determination of Cadmium, Pb, Cu, Zn, and Se in human blood of jordanian smokers by ICP-OES. *Biological Trace Element Research*, 133(1), 1-11.

Mutti, A., Corradi, M., Goldoni, M., Vettori, M.V., Bernard, A., Apostoli, P. (2006). Exhaled metallic elements and serum pneumoproteins in asymptomatic smokers and patients with COPD or asthma. *Chest*. 129 (5):1288-1297.

Nordberg, G. F., Kjellstrom, T., Nordberg, M. (1985). Kinetics and metabolism. In Friberg, L. (Ed.) *Cadmium and health: A toxicological and epidemiological appraisal. Exposure, Dose and Metabolism*. p.p. 103-178. CRC Press.

Olsson, I. M., Bensryd, I., Lundh, T., Ottosson, H., Skerfving, S., Oskarsson, A. (2002). Cadmium in blood and urine-impact of sex, age, dietary intake, iron status, and former smoking association of renal effects. *Environmental Health Perspectives*, 110(12), 1185-1190.

Osredkar, J., Sustar, N. (2011). Copper and Zinc. Biological Role and Significance of Copper/Zinc Imbalance. *Journal of Clinical Toxicology*, S3:001, 1-8.

Prashanth L., Kattapagari, K. K., Chitturi, R. T., Baddam, V. R. R., Prasad, L. K. (2015) A Review on Role of Essential Trace Elements in Health and Disease. *Journal of Dr. NTR University of Health Sciences*, 4(2), 75-85.

Proudfoot, C., McPherson, A. L., Kolb, A. F., Stark, W. M. (2011). Zinc finger recombinases with adaptable DNA sequence specificity. *PloS one*, 6(4), e19537.

Richter, P., Faroon, O., Pappas, R. S. (2017). Cadmium and Cadmium/Zinc Ratios and Tobacco-Related Morbidities. *International Journal of Environmental Research and Public Health*, 14(10), 1154.

Talhout, R., Schulz, T., Florek, E., van Benthem, J., Wester P., Opperhuizen, A. (2011) Hazardous Compounds in Tobacco Smoke. *International Journal of Environmental Research and Public Health*, *8*, 613-628.

Waisberg, M., Joseph, P., Hale, B., Beyersmann, D. (2003) Molecular and cellular mechanisms of cadmium carcinogenesis. *Toxicology*, 192(2-3), 95-117.

Summary/Sažetak

Istraživanja su pokazala da pušenje cigareta utiče na akumulaciju nekih teških metala u pojedinim tkivima i na metabolizam esencijalnih elemenata. Cilj istraživanja je bio utvrditi razlike koncentracija kadmijuma u krvi i urinu te cinka u urinu pušača i bivših pušača u odnosu na nepušače i utvrditi mogući uticaj koncentracije kadmijuma na ekskreciju esencijalnog elementa cinka. Istraživanjem je obuhvaćeno 106 osoba. Ispitanici su bili stalni pušači (n=51), bivši pušači (n=38) i nepušači (n=17). Za određivanje kadmijuma korištena je metoda atomske apsorpcione spektrofotometrije (AAS) sa elektrotermalnim atomizerom. Cink je određen metodom ASS sa plamenim atomizerom. Utvrđena je značajna razlika u vrijednostima kadmijuma u krvi između skupina: pušači i nepušači ($p<0.001$), pušači i bivši pušači ($p<0.001$), te između bivših pušača i nepušača ($p=0,045$). Postoji značajna pozitivna i jaka korelacija u nivou cinka i kadmijuma u urinu na gram kreatinina, te povećanjem nivoa kadmijuma povećava se i nivo cinka ($rho= 0.781$; $p= 0.001$). Rezultati naše studije pokazuju da je pušenje faktor koji može povećati nivo kadmijuma u mjeri koja će znatno povećati ekskreciju cinka, tj. njegov povećan gubitak.

In memoriam

Mustafa Memić
(1963-2020)

Mustafa Memić, rođen je 18.02.1963. u Sjenici, gdje je završio osnovno i srednje obrazovanje. *Zvanje diplomiranog inženjera hemije stekao je na Odsjeku za hemiju Prirodno-matematičkog fakulteta Univerziteta u Sarajevu 1989. godine, gdje je naredne godine izabran za asistenta. Postdiplomski studij je okončao 2001. godine kao i doktorat hemijskih nauka na Prirodno-matematičkom fakultetu u Sarajevu, 2007. godine.*

Svoj radni vijek je proveo na Katedri za analitičku hemiju Odsjeka za hemiju Prirodno-matematičkog fakulteta. U zvanje redovnog profesora je izabran 2016. godine. Na Prirodno-matematičkom fakultetu nije bilo samo njegovo radno mjesto, to je od prvih dana njegovog zaposlenja bio njegov drugi dom. Domaćinski, sa puno brige i poštenoga rada je obavljao svoje poslove. Bio omiljen među saradnicima i studentima.

Imao je bogato nastavno-pedagoško iskustvo u radu na više fakulteta Univerziteta u Sarajevu. Posebno uspješne rezultate pokazao je u radu sa studentima Prirodno-matematičkog fakulteta u realizaciji nastave iz većeg broja predmeta u oblasti analitičke hemije. Pod mentorstvom prof. Memića urađen je veliki broj magistarskih radova i završnih radova drugog ciklusa studija, kao i završnih radova prvog ciklusa studija i diplomskih radova. Bio je mentor na dvije uspješno odbranjene doktorske disertacije, a još dvije disertacije je doveo skoro do kraja.

Bio je šef Katedre za analitičku hemiju, šef Odsjeka za hemiju, prodekan za nastavu i naučnoistraživački rad Prirodno-matematičkog fakulteta i od kraja 2016. godine dekan Prirodno-matematičkog fakulteta. Također, bio je aktivni član Društva hemičara i tehnologa Kantona Sarajevo, član redakcijskog odbora „Glasnika hemičara i tehnologa BiH“, saradnik iz oblasti mjeriteljstva u Institutu za mjeriteljstvo BiH, član radne grupe za okolinu u Tehničkom komitetu Instituta za standardizaciju BiH, saradnik Instituta za akreditaciju BiH u svojstvu eksperta-ocjenjivača, predsjednik Skupštine Društva hemičara i tehnologa Kantona Sarajevo, voditelj postdiplomskog studija na Odsjeku za hemiju

Prirodno-matematičkog fakulteta, te stručni saradnik u Centru za istraživanje i razvoj novih metala. Dao je veliki doprinos odbrani Bosne i Hercegovine.

Objavio je veliki broj originalnih naučnih radova u priznatim publikacijama, te učestvovao na više međunarodnih naučnih i stručnih skupova na kojima je kao autor i koautor predstavio veliki broj radova čiji su sažeci objavljeni u zbornicima. Kao koautor i autor napisao je tri recenzirana univerzitetska udžbenika, te učestvovao u realizaciji više međunarodnih i domaćih naučno-istraživačkih projekata.

Živio je životom poštenoga čovjeka. Izuzetan hemičar i sjajan nastavnik, prof. Memić je ostao takav do kraja svog radnog vijeka i prerane smrti. Bio je snažan autoritet ali pri tome skroman i velikodušan čovjek, izuzetno cijenjen među studentima i kolegama. Zbog izuzetno plodnog naučnoga doprinosa te nesebičnog zalaganja za dobrobit Prirodno-matematičkog fakulteta ostat će nam zadatak da se sa puno zahvalnosti sjećamo našeg Mustafe Memića.

INSTRUCTIONS FOR AUTHORS

GENERAL INFORMATION

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina (Glasnik hemičara i tehnologa Bosne i Hercegovine) is a semiannual international journal publishing papers from all fields of chemistry and related disciplines.

Categories of Contributions

1. *Original Scientific Papers* – (about 10 typewritten pages) report original research which has not been published previously, except in a preliminary form. The paper should contain all the necessary information to enable reproducibility of the described work.
2. *Short Communications* – (about 5 typewritten pages) describing work that may be of a preliminary nature but which merits immediate publication.
3. *Notes* – (about 3 typewritten pages) report unpublished results of short, but complete, original research or describe original laboratory techniques.
4. *Reviews* – (about 30 typewritten pages) present a concise and critical survey of a specific research area. Generally, these are prepared by the invitation of the Editor.
5. *Book and Web Site Reviews* – (about 2 typewritten pages).
6. *Extended Abstracts* – (about 2 typewritten pages) of Lectures given at international meetings.

Reviewing the Manuscript

All contributors are evaluated according to the criteria of originality and quality of their scientific content, and only those deemed worthy will be accepted for publication. To facilitate the reviewing process, authors are encouraged to suggest three persons competent to review their manuscript. Such suggestions will be taken into consideration but not always accepted.

The Editor-In-Chief and Editors have the right to decline formal review of a manuscript when it is deemed that the manuscript is:

1. on a topic outside the scope of the Journal;
2. lacking technical merit;
3. of insufficient novelty for a wide international readership;
4. fragmentary and providing marginally incremental results; or
5. is poorly written.

Proofs

When a manuscript is ready for printing, the corresponding author will receive a PDF-formatted manuscript for proof reading, which should be returned to the journal within one week. Failure to do so will be taken as the authors are in agreement with any alteration which may have occurred during the preparation of the manuscript.

Copyright

Subscribers may reproduce tables of contents or prepare lists of articles including abstracts for internal circulation within their institutions. Permission of the Publisher is required for resale or distribution outside the institution and for all other derivative works, including compilations and translations.

Professional Ethics and Publication Policy

The journal expects the Editors, Referees and authors to adhere to the well-known standards of professional ethics. Authors are responsible for the factual accuracy of their contributions. Submission of the paper commits the author not to submit the same material elsewhere. Referees should act promptly. If certain circumstances preclude prompt attention to the manuscript at the time it is received, the non-received manuscript should be returned immediately to the Editor or the Referee should contact the Editor for possible delay of the report submission date. The Editor accepts full responsibility for his decisions on the manuscripts.

PREPARATION AND SUBMISSION OF MANUSCRIPT

Cover Letter

Manuscripts must be accompanied by a cover letter in which the type of the submitted manuscript. It should contain:

1. full name(s) of the author(s),
2. mailing address (address, phone and fax numbers, e-mail) of the author to whom correspondence should be addressed,
3. title of the paper (concise, without any abbreviations),
4. type of contribution,
5. a statement that the article is original and is currently not under consideration by any other journal or any other medium, including preprints, electronic journals and computer databases in the public domain, and
6. the names, full affiliation (department, institution, city and country), and
7. e-mail addresses of three potential Referees.

Contributors from Bosnia and Herzegovina should provide the name and full affiliation of at least one Referee from abroad.

Authors are fully encouraged to use ***Cover Letter Template*****.**

Manuscript preparation

The submitted articles must be prepared with Word for Windows. Manuscripts should be typed in English (either standard British or American English, but consistent throughout) with 1.5 spacing (12 points Times New Roman; Greek letters in the character font Symbol) in A4 format leaving 2.5 cm for margins. Authors are fully encouraged to use ***Manuscript Template***.

All contributions should be written in a style that addresses a wider audience than papers in more specialized journals. Manuscripts with grammar or vocabulary deficiencies are disadvantaged during the scientific review process and, even if accepted, may be returned to the author to be rewritten in idiomatic English. The authors are requested to seek the assistance of competent English language expert, if necessary, to ensure their English is of a reasonable standard. The journal maintains its policy and takes the liberty of correcting the English of manuscripts scientifically accepted for publication.

Tables and figures and/or schemes should not be embedded in the manuscript but their position in the text indicated. In electronic version (Word.doc document) tables and figures and/or schemes should follow the text, each on a separate page. Please number all pages of the manuscript including separate lists of references, tables and figures with their captions.

IUPAC and International Union of Biochemistry and Molecular Biology recommendations for the naming of compounds should be followed.

SI units, or other permissible units, should be employed. The designation of physical quantities should be in Times New Roman font. In text, graphs, and tables, brackets should be used to separate the designation of a physical quantity from the unit. Please do not use the axes of graphs for additional explanations; these should be mentioned in the figure captions and/or the manuscript (example: “pressure at the inlet of the system, kPa” should be avoided).

Percents and *per mills*, although not being units in the same sense as the units of dimensioned quantities, can be treated as such. Unit symbols should never be modified (for instance: w/w %, vol.%, mol.%) but the quantity measured has to be named, *e.g.* mass fraction, w=95 %; amount (mole) fraction, x=20 %.

Latin words, as well as the names of species, should be in *italic*, as for example: *i.e., e.g., in vivo, ibid, Artemisia annua* L., *etc*. The branching of organic compound should also be indicated in *italic*, for example, *n*-butanol, *tert*-butanol, *etc*.

Decimal numbers must have decimal points and not commas in the text (except in the Bosnian/Croatian/Serbian abstract), tables and axis labels in graphical presentations of results. Thousands are separated, if at all, by a comma and not a point.

Structure of the Manuscript

The manuscript must contain, each on a separate page, the title page, abstract in English, (abstract in Bosnian/Croatian/Serbian), graphical abstract (optional), main text, list of references, tables (each table separately), illustrations (each separately), and legends to illustrations (all on the same page).

1. ***Title page*** must contain: the title of the paper (bold letters), full name(s) of the author(s), full mailing addresses of all authors (italic), keywords (up to 6), the phone and fax numbers and the e-mail address of the corresponding author.
2. A one-paragraph ***abstract*** written of 150–200 words in an impersonal form indicating the aims of the work, the main results and conclusions should be given and clearly set off from the text. Domestic authors should also submit, on a separate page, a Summary/Sažetak. For authors outside Bosnia and Herzegovina, the Editorial Board will provide a Bosnian/Croatian/Serbian translation of their English abstract.
3. Authors are encouraged to submit a ***graphical abstract*** that describes the subject matter of the paper. It should contain the title of the paper, full name(s) of the author(s), and graphic that should be no larger than 11 cm wide by 5 cm tall. Authors are fully encouraged to use ***Graphical Abstract Template.***
4. ***Main text*** should have the following form:
   - ***Introduction***should include the aim of the research and a concise description of background information and related studies directly connected to the paper.
   - ***Experimental*** section should give the purity and source of all employed materials, as well as details of the instruments used. The employed methods should be described in sufficient detail to enable experienced persons to repeat them. Standard procedures should be referenced and only modifications described in detail.
   - ***Results and Discussion***should include concisely presented results and their significance discussed and compared to relevant literature data. The results and discussion may be combined or kept separate.
   - The inclusion of a ***Conclusion***section, which briefly summarizes the principal conclusions, is highly recommended.
   - ***Acknowledgement*** (optional).
   - Please ensure that every ***reference*** cited in the text is also present in the reference list (and *vice versa*). Unpublished results and personal communications are not recommended in the reference list, but may be mentioned in the text. If these references are included in the reference list they should follow the standard reference style of the journal and should include a substitution of the publication date with either "Unpublished results" or "Personal communication" Citation of a reference as "in press" implies that the item has been accepted for publication. As a minimum, the full URL should be given and the date when the reference was last accessed. Any further information, if known (DOI, author names, dates, reference to a source publication, etc.), should also be given. No more than 30 references should be cited in your manuscript.

     In the text refer to the author's name (without initials) and year of publication (e.g. "Steventon, Donald and Gladden (1994) studied the effects..." or "...similar to values reported by others (Anderson, Douglas, Morrison, *et al.*, 1990)..."). Type the names of the first three authors at first citation. At subsequent citations use first author *et al.* The list of references should be arranged alphabetically by

authors' names and should be as full as possible, listing all authors, the full title of articles and journals, publisher and year.

Examples of ***reference style***:

a) Reference to a journal publication:
   Warren, J. J., Tronic, T. A., Mayer, J. M. (2010). Termochemistry of proton-coupled electron transfer reagents and its implications. *Chemical Reviews*, *110 (12)*, 6961-7001.
b) Reference to a book:
   Corey, E. J., Kurti, L. (2010). *Enantioselective chemical synthesis*. (1st Ed.) Direct Book Publishing, LLC.
c) Reference to a chapter in an edited book:
   Moody, J. R., Beck II, C. M. (1997). Sample preparation in analytical chemistry. In Setlle, F. A. (Ed.), *Handbook of instrumental techniques for analytical chemistry*. (p.p. 55-72). Prentice Hall.
d) Reference to a proceeding:
   Seliskar, C. J., Heineman, W.R., Shi, Y., Slaterbeck, A.F., Aryal, S., Ridgway, T.H., Nevin, J.H. (1997). *New spectroelectrochemical sensor*, in Proceedings of 37th Conference of Analytical Chemistry in Energy and Technology, Gatlinburg, Tenesee, USA, p.p. 8-11.
e) Patents:
   Healey, P.J., Wright, S.M., Viltro, L.J., (2004).*Method and apparatus for the selection of oral care chemistry*, The Procter & Gamble Company Intellectual Property Division, (No.US 2004/0018475 A1).
f) Chemical Abstracts:
   Habeger, C. F., Linhart, R. V., Adair, J. H. (1995). Adhesion to model surfaces in a flow through system. *Chemical Abstracts,* CA 124:25135.
g) Standards:
   ISO 4790:1992. (2008). *Glass-to-glass sealings - Determination of stresses*.
h) Websites:
   Chemical Abstract Service, www.cas.org, (18/12/2010).

- ***Tables*** are part of the text but must be given on separate pages, together with their captions. The tables should be numbered consequently in Latin numbers. Quantities should be separated from units by brackets. Footnotes to tables, in size 10 font, are to be indicated consequently (line-by-line) in superscript letters. Tables should be prepared with the aid of the Word table function, without vertical lines. Table columns must not be formatted using multiple spaces. Table rows must not be formatted using Carriage returns (enter key; ↵ key). Tables should not be incorporated as graphical objects.
- ***Figures and/or Schemes*** (in high resolution) should follow the captions, each on a separate page of the manuscript. High resolution illustrations in TIF or EPS format (JPG format is acceptable for colour and greyscale photos, only) must be uploaded as a separate archived (.zip or .rar) file.

Figures and/or Schemes should be prepared according to the artwork instructions.

- ***Mathematical and chemical equations*** must be numbered, Arabic numbers, consecutively in parenthesis at the end of the line. All equations should be embedded in the text except when they contain graphical elements (tables, figures, schemes and formulae). Complex equations (fractions, inegrals, matrix…) should be prepared with the aid of the Word Equation editor.

Artwork Instructions

Journal accepts only TIF or EPS formats, as well as JPEG format (only for colour and greyscale photographs) for electronic artwork and graphic files. MS files (Word, PowerPoint, Excel, Visio) are NOT acceptable. Generally, scanned instrument data sheets should be avoided. Authors are responsible for the quality of their submitted artwork.

Image quality: keep figures as simple as possible for clarity - avoid unnecessary complexity, colouring and excessive detail. Images should be of sufficient quality for the printed version, i.e. 300 dpi minimum.

Image size: illustrations should be submitted at its *final size* (8 cm for single column width or 17 cm for double column width) so that neither reduction nor enlargement is required.

Photographs: please provide either high quality digital images (250 dpi resolution) or original prints. Computer print-outs or photocopies will not reproduce well enough for publication. Colour photographs rarely reproduce satisfactorily in black and white.

The facility exist for color reproduction, however the inclusion of color photographs in a paper must be agreed with Editor in advance.

Reporting analytical and spectral data

The following is the recommended style for analytical and spectral data presentation:

1. ***Melting and boiling points***:
   mp 163–165°C (lit. 166°C)
   mp 180°C dec.
   bp 98°C
   Abbreviations: mp, melting point; bp, boiling point; lit., literature value; dec, decomposition.
2. ***Specific Rotation***:
   $[a]^{23}_{D}$ –222 (*c* 0.35, MeOH).
   Abbreviations: a, specific rotation; D, the sodium D line or wavelength of light used for determination; the superscript number, temperature (°C) at which the determination was made; In parentheses: *c* stands for concentration; the number following *c* is the concentration in grams per 100 mL; followed by the solvent name or formula.

3. ***NMR Spectroscopy***:
   ^{1}H NMR (500 MHz, DMSO-d_6) d 0.85 (s, 3H, CH_3), 1.28–1.65 (m, 8H, 4´CH_2), 4.36–4.55 (m, 2H, H-1 and H-2), 7.41 (d, *J* 8.2 Hz, 1H, ArH), 7.76 (dd, *J* 6.0, 8.2 Hz, 1H, H-1'), 8.09 (br s, 1H, NH).
   ^{13}C NMR (125 MHz, $CDCl_3$) d 12.0, 14.4, 23.7, 26.0, 30.2, 32.5, 40.6 (C-3), 47.4 (C-2'), 79.9, 82.1, 120.0 (C-7), 123.7 (C-5), 126.2 (C-4).
   Abbreviations: d, chemical shift in parts per million (ppm) downfield from the standard; *J*, coupling constant in hertz; multiplicities s, singlet; d, doublet; t, triplet; q, quartet; and br, broadened. Detailed peak assignments should not be made unless these are supported by definitive experiments such as isotopic labelling, DEPT, or two-dimensional NMR experiments.
4. ***IR Spectroscopy***:
   IR (KBr) n 3236, 2957, 2924, 1666, 1528, 1348, 1097, 743 cm^{-1}.
   Abbreviation: n, wavenumber of maximum absorption peaks in reciprocal centimetres.
5. ***Mass Spectrometry***:
   MS *m/z* (relative intensity): 305 (M^+H, 100), 128 (25).
   HRMS–FAB (*m/z*): $[M+H]^+$calcd for $C_{21}H_{38}N_4O_6$, 442.2791; found, 442.2782.
   Abbreviations: *m/z*, mass-to-charge ratio; M, molecular weight of the molecule itself; M^+, molecular ion; HRMS, high-resolution mass spectrometry; FAB, fast atom bombardment.
6. ***UV–Visible Spectroscopy***:
   UV (CH_3OH) l_{max} (log e) 220 (3.10), 425 nm (3.26).
   Abbreviations: l_{max}, wavelength of maximum absorption in nanometres; e, extinction coefficient.
7. ***Quantitative analysis***:
   Anal.calcd for $C_{17}H_{24}N_2O_3$: C 67.08, H 7.95, N 9.20. Found: C 66.82, H 7.83, N 9.16.All values are given in percentages.
8. ***Enzymes and catalytic proteins relevant data:***
   Papers reporting enzymes and catalytic proteins relevant data should include the identity of the enzymes/proteins, preparation and criteria of purity, assay conditions, methodology, activity, and any other information relevant to judging the reproducibility of the results[1]. For more details check Beilstein Institut/STRENDA (standards for reporting enzymology data) commission Web site (http://www.strenda.org/documents.html).

Submission Checklist

The following list will be useful during the final checking of an article prior to sending it to the journal
for review:

- E-mail address for corresponding author,

[1] For all other data presentation not mentioned above please contact Editor for instructions.

- Full postal address,
- Telephone and fax numbers,
- All figure captions,
- All tables (including title, description, footnotes),
- Manuscript has been "spellchecked" and "grammar-checked",
- References are in the correct format for the journal,
- All references mentioned in the Reference list are cited in the text, and *vice versa*.

Submissions

Submissions should be directed to the Editor by e-mail: ***glasnik@pmf.unsa.ba***, or ***glasnikhtbh@gmail.com***. All manuscripts will be acknowledged on receipt (by e-mail) and given a reference number, which should be quoted in all subsequent correspondence.

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

Print ISSN: 0367-4444
Online ISSN: 2232-7266

Zmaja od Bosne 33-35, BA-Sarajevo
Bosnia and Herzegovina
Phone: +387-33-279-918
Fax: +387-33-649-359
E-mail: glasnik@pmf.unsa.ba
glasnikhtbh@gmail.com

Sponsors

Glasnik hemičara i tehnologa Bosne i Hercegovine

Bulletin of the Chemists and Technologists of Bosnia and Herzegovina

Print ISSN: 0367-4444
Online ISSN: 2232-7266

Zmaja od Bosne 33-35, BA-Sarajevo
Bosnia and Herzegovina
Phone: +387-33-279-918
Fax: +387-33-649-359
E-mail: glasnik@pmf.unsa.ba
glasnikhtbh@gmail.com

www.elektroprivreda.ba/stranica/te-kakanj

FMON
Federalno ministarstvo obrazovanja i nauke

www.fmon.gov.ba

Ministarstvo za obrazovanje, nauku i mlade Kantona Sarajevo

https://mon.ks.gov.ba